

SOME EFFECTS OF PLANT GROWTH SUBSTANCES
ON BROAD BEANS (*VICIA FABA* L. *MAJOR*)

PAUL D. RYLOTT



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DECLARATION

This is to declare that this thesis has
out herein was also carried out
by myself, except where specifically stated.

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Abstract

Yield fluctuation in *Vicia faba* is due primarily to reproductive failure, which can occur as a result of bud abortion, flower shedding or pod and ovule abortion (Gates *et al.*, 1983b). Flower drop, which accounts for the major proportion of total reproductive loss, contributes most to the reduction in potential yield. Application of artificial plant growth retardants (EL500, JF 10405 and Alar) were shown to increase the yield of broad beans (*Vicia faba* L. *major* cv. Threefold White), by up to 52%, mostly due to increased pod set.

Experiments involving the application of plant growth substances directly to the flowers, suggested that increase in pod set was due to changes in intrinsic hormone levels. In particular, high levels of cytokinin are required at the pedicel:peduncle junction pre-pollination, to allow successful initiation of potential sinks, while increased levels of auxin are required after pollination in conjunction with cytokinin to allow cell division, pod expansion and vascular differentiation. Application of anti-gibberellin plant growth retardants appeared to alter internal hormone ratios, affecting the distribution of dry matter production during early flowering, i.e. treated plants had an increased root to stem dry matter growth rate. This suggested an increased cytokinin:gibberellin ratio had been achieved.

Although it was shown that pod set could be enhanced by the application of either plant growth retardants and/or plant growth substances, yield was not always as high as anticipated due to increased levels of pod drop. Further applications of cytokinin and auxin to the pods reduced this drop. It followed therefore, that further applications could dramatically increase the yield potential of the plant due to better distribution of assimilates to pods. However, it would appear that the plant also suffers from source limitations and until these are successfully overcome, yield instability in the field environment is still likely.

Acknowledgements

I would expressly like to thank my supervisor, Dr. M.L. Smith for his valuable advice and constructive criticism during the preparation of this thesis. Similarly, I would also like to thank my second supervisor Dr. J.H. Lennard and Mr. E.A. Hunter at SASS for help with statistics. Thanks also to Robert Redpath and his "merry men", especially, Jeanette, Brian, Susan and Dan at the Bush Estate, whose help during field trials was greatly appreciated. I would also like to acknowledge the financial support of a MAFF scholarship.

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Last and definitely not least, thanks Tan.

Chapter 1

Literature Review

Introduction

The broad bean (*Vicia faba* L. *major*), is an annual plant of indeterminate growth habit, bearing creamy-white flowers on axillary racemes, which typically develop after an average of 5 vegetative nodes have been formed. The number of flower bearing nodes is typically 10 - 12 but can range from 8 - 16. Broad beans are one of the oldest cultivated crops, thought to originate from central Asia (Ladizinski, 1975). The crop was very popular with ancient Egyptians and Romans and due to its high protein content and easiness of drying, was so important in the UK during the middle ages that to steal broad beans from an open field carried the death penalty (Anon., 1984). The crops popularity has declined in recent years, partly because of imported protein beans such as soya (*Glycine max*) and navy bean (*Phaseolus vulgaris*), however, within the U.K. between 3511 (1982) and 6380 (1979), hectares are sown every year, which represents approx. 2% of the vegetable acreage (Anon., 1984).

The crop is grown for three major markets, freezing, canning and the fresh market. Approximately 75% of the crop is grown for processing and the main cropping areas are consequently close to processing plants in Lincolnshire and Norfolk. Broad beans for the fresh market are more typically grown in the market garden areas of Kent, Hereford and Worcestershire.

As with the crop's sister sub-species, the field bean (*Vicia faba* L. *minor*), the crop has a tendency to suffer from yield fluctuations. Field beans have yielded 7 - 9t /ha (Sprent *et al.*, 1977), but average yields are closer to 3t/ha (Anon., 1981). Hawtin and Hebblethwaite (1983), suggest that the yield stability index for faba bean is 137% compared to 53% for wheat. As could be expected, figures suggest a similar phenomena occurs in broad beans, with yields ranging between 10.4 t/ha in 1972 and 3.2 t/ha in 1976 (Anon., 1983). Although the crop offers a valuable rotational

break from cereals, this yield instability coupled with its pricing instability has added to the reduction in its popularity with growers.

Yield Instability

Yield instability problems can be split into two major categories, extrinsic and intrinsic. Extrinsic factors involve agronomy such as weeds, disease and pest problems (which are now controllable in the main due to modern pesticides) and pollination.

The faba bean is both self and cross fertilised, with the frequency of cross fertilisation ranging from 5 - 80%, however, it is more typically in the range of 30-40% (Holden and Bond, 1960). Much of the reason for the range can be attributed to fluctuations in local bee populations (Free and Williams, 1976). Consequently, the conversion of the crop to total autogamy would be a contributory factor to yield stability, provided inbreeding depression could be avoided (Lawes, 1974; Hawtin, 1981). Kambal *et al.* (1976), supported such a view, but added that as all pollinated flowers did not develop into mature pods, factors other than pollination must also be important.

Reproductive loss which can occur as bud abortion, flower drop, pod drop or ovule abortion, has been shown to be the major factor in yield instability of faba beans (Smith, 1982), with much of this loss due to intra-plant competition. The abortion of buds due primarily to competition for assimilates, is common in the crop, but is of less significance than other reproductive losses (Gates *et al.*, 1983b). Internal competition for assimilates is also cited as a contributory factor leading to pod drop. Gehringer and Keller (1980), stated that this was accentuated by a loss of photosynthetic area. Smith (1982) further demonstrated the competitive effects by removing lower racemes, resulting in greater pod retention at upper racemes. Inter-ovule competition for assimilates generated by those nearest to the stigma having a

slight developmental advantage is cited as one reason for this loss (Gates *et al.*, 1983b). Flower drop, which accounts for the major proportion of reproductive loss, contributes most to reduction in potential yield (Kambal, 1969; Chapman and Peat, 1978; Smith, 1982). Estimates of flower loss range from 48 - 97% (Soper, 1952; Kambal, 1969; Jaquiere and Keller, 1978; Smith, 1982). Chapman *et al.* (1979), concluded that any treatment that predisposed that plant to increased competition increased flower drop. Thus a range of environmental conditions such as: water supply (El-Rahman *et al.*, 1980), density (Sprent *et al.*, 1977) and light (Hodgeson and Blackman, 1957; Smith, 1982); which all limit assimilate supply and hence increase competition have been cited as factors affecting flower abscission.

Flower drop within *Vicia faba*, follows a characteristic pattern. The incidence of shedding at lower racemes and proximal flower positions within the raceme is low and stable, whereas at more apical racemes and distal positions within those racemes is higher and more variable (Gates *et al.* 1983b). Within individual racemes, the pattern of flower drop can be related to acropetal anthesis (Gates *et al.*, 1981) and the branched nature of the vascular system within the raceme (Gates *et al.*, 1983b). Pollination of the earliest flower to reach anthesis, stimulates physiological changes leading to flower abscission at distal sites. Selection of inbred lines that showed synchrony of flowering, were shown to exhibit complete flower retention (Smith, 1982). In addition, in conventional varieties, removal of the proximal two or three flowers on the raceme, reduced flower drop at distal positions (Smith, 1982). This suggests that an abscission promoting substance is translocated from young pods to distal buds (Gates *et al.*, 1983a).

Such findings have obviously lead to experimentation with plant growth substances in an attempt to alleviate this problem.

Plant Growth Substances

Plant growth substances can be divided into two categories, plant hormones and artificial plant growth regulators. Both can be described as organic substances, other than nutrients, which, when present in small amounts can evoke specific biochemical, physiological or morphological responses (Moore, 1979). All hormones are growth regulators, but the converse is not true (Keller and Belluci, 1983). Plant hormones are divided into five groups: auxins, gibberellins, cytokinins, inhibitors (abscisic acid) and ethylene.

Auxins

The auxin, indole acetic acid (IAA), was the first naturally occurring plant growth substance to be isolated. It was later found, together with its derivatives to be present in most plants and is considered to be the principal true auxin of higher plants. Its isolation allowed synthesis of artificial auxins such as indole-3-butyric acid (IBA), 1-naphthylacetic acid (NAA) and 2,4-dichlorophenoxyacetic acid (2,4-D). Auxins are known to cause many different effects in the same plant, examples of these responses include: stimulation of root growth, control of vascular system differentiation, control of tissue culture differentiation, control of apical dominance, delay of senescence, promotion of flowering, fruit setting and ripening (Bandruski and Nonhebel, 1984). The chlorinated auxin, 4-chloroindole -3- acetic acid has been shown to occur in *Pisum* and *Vicia spp.* (Engvild *et al.*, 1980). Hoffinger and Bottger (1979), demonstrated that this auxin was 10 times more active than IAA in the *Avena* straight growth assay.

Gibberellins

Gibberellins were discovered shortly after auxins, but it was not until 1954, that a pure gibberellin (GA₃), was isolated and its chemical structure determined. Since then many more have been discovered. Indeed, Jones and MacMillan (1984), suggest that there are 62 individual gibberellins, all having the same gibbane

skeleton. In general, immature seeds contain more gibberellins than vegetative tissue, hence most discoveries have been made from seeds. Two major routes of research have emphasised the role of gibberellins in RNA and protein synthesis, while the other has concentrated on the growth rate of stems and leaves due to an alteration in cell expansion and or division (Jones and MacMillan, 1984). GAs also appear to play a role in flowering, with levels increasing prior to anthesis, suggesting a role in anthesis and or flower opening (Pharis and King, 1985).

Cytokinins

A cytokinin is described as a compound, which in the presence of optimal auxin, induces cell division in the tobacco pith or similar assay, grown on an optimally defined medium (Horgan, 1984). The first naturally occurring cytokinin (zeatin), was isolated from immature kernels of *Zea mays* (Letham, 1963). Synthetic cytokinins include: kinetin, benzyladenine (BA) and 6-benzylaminopurine (BAP) and substituted ureas (Garrod, 1982).

As well as being active in cell division, cytokinins can also increase the size of leaves or cotyledons by cell expansion and delay chlorophyll and protein degradation (Horgan, 1984). This anti-senescent effect is related to their ability to divert numerous substances to areas that have been treated with them. Horgan (1984), suggests the reason for this is the increased rate of metabolism in these treated sites.

Abscisic acid

Van Stevenik (1959), found that the abortion of fertilized yellow lupin (*Lupinus luteus*), pods was correlated with the amount of inhibitory material present. This work was pursued by Rothwell and Wain (1964), who extracted an active fraction, from which abscisic acid (ABA) was later isolated (Cornforth *et al.*, 1966). ABA

has been found in all higher plant material examined including: roots, xylem sap, phloem sap, pollen, petals, fruits and seeds (Milborrow, 1984).

ABA has a wide range of physiological effects. In cotton explants it has been shown to stimulate abscission of petiolar stumps (Osborne *et al.*, 1972), although its effect is much reduced in intact plants. It has also been shown to be correlated with dormant potato tubers, the ripening of fruit and the rapid closure of stomata in leaves (Milborrow, 1984).

Ethylene

At the turn of the century, physiologists were aware that a component of coal gas was a potent promoter of abscission, this was later discovered to be ethylene (Nejubow, 1901). It is also involved in: control of apical dominance, bud growth inhibition, flower inhibition, latex flow promotion, fruit ripening in bananas and tomatoes and root growth inhibition (Abeles, 1985).

Synthetic Plant Growth Regulators

Synthetic plant growth regulators include: growth retardants, growth inhibitors, hormone transport inhibitors and ethylene promoters or inhibitors.

Plant Growth Inhibitors

Cathey (1964), defined a growth inhibitor as a substance which completely inhibits growth in the shoot meristems and at high concentrations, suppresses all growth. Maleic hydrazide was the first synthetic plant growth inhibitor to gain widespread commercial usage. It is used for the prevention of sprouting of potatoes and onions during storage (Garrod, 1982) and is also used for the suppression of grass verges (Dicks, 1976). Another similar product is tecnazene, which is also used as a sprout suppressant in potatoes (Ivens, 1990).

Ethylene promoters / inhibitors

The use of ethylene in agriculture was hampered for several years because it was a gas. This was eased, however, with the advent of ethephon (Karbachnik and Rossiyskava, 1946), ^{which} was not patented until 1965. Below pH 4.1, ethephon is chemically stable, but on entering a plant (which is less acid) liberates ethylene (Garrod, 1982). Today, ethephon, or ethephon plus mepiquat chloride is used as a straw shortener and "anti-brackling" agent in barley and rye (Ivens, 1990).

Ethylene inhibitors can be sub-divided into conversion inhibitors and receptor inhibitors. Conversion inhibitors prevent the precursor, methionine, being converted into ethylene and include rhizobitacin and canaline (Abeles, 1985).

Receptor inhibitors cause a reaction at the receptor site, that substitutes copper ions for silver ions, so preventing the action of ethylene (Beyer, 1976). Silver ions are added in the form of silver nitrate or silver thiosulphate (Veen, 1979).

Hormone Transport Inhibitors

These substances act to block the movement of endogenous hormones, which can in turn lead to local accumulations of these hormones and hence physiological change. The plant growth regulator TIBA (2,3,5-triiodobenzoic acid), is thought to act in this way by blocking the downward movement of IAA from the stem apex (Luckwill, 1981).

TIBA was patented in 1961 as a product for the enhancement of flower bud formation and widening branch angle in apples (Garrod, 1982). It was also shown to increase yields in soybeans (Greer and Anderson, 1965), due to increases in the number of pods per plant at final harvest (Boize, 1982). In *Vicia faba*, trials have shown that TIBA caused a reduction in plant height and increased the number of tillers and pods (El-Zawily *et al.*, 1985). Yield increases of 10% were also described (Attiya *et al.*, 1983), after application to field beans at the 6 leaf stage

and the commencement of flowering. Newaz and Lawes (1980) also demonstrated that the application of TIBA could lead to increases in faba bean yields of up to 7%.

Morphactins, which are derivatives of fluorene-9-carboxylic acid, were reported by Schneider *et al.* (1965) to induce a gradual inhibition of growth often accompanied by stimulation of branching. As with TIBA, this effect is due to the interruption of downward movement of auxins from the plant apex (Garrod, 1982). Application of the morphactin CME 73170P (2-chloro-9-hydroxyfluoren-9-carboxylic acid), at levels of 1 - 10 mg/ litre, increased the yield of *Vicia faba* by up to 58% (El-Zawily *et al.*, 1985). Yield increases were ascribed to an increase in the number of tillers and pods retained, with the later possibly due to decreased levels of ABA.

Growth Retardants

Growth retardants are synthetic organic chemicals, which when applied to responsive plants, reduce the rate of stem elongation by inhibiting sub-apical meristem activity, normally without affecting leaf production and development or inducing growth malformations (Cathey, 1964). It is almost always possible to reverse the effect of a retardant on stem elongation by a later application of gibberellic acid (Dicks, 1976).

Chlormequat

A series of quaternary ammonium compounds showing growth retarding properties was reported in 1960, by Tolbert, of which the most common is chlormequat (2-chloroethyl trimethylammonium chloride). Its main effect is a reduction in plant height caused by shortening basal internodes, especially in wheat and oats, thereby reducing lodging losses under conditions of high fertility (Humphries, 1968). Yields of wheat can also be increased in the absence of lodging due to increases in the number of grains per ear and fertile tillers per unit area (Huntley-Bull and Schwabe, 1982).

Chlormequat applied to *Vicia faba* plants (Hassan and El-Moursi, 1982) increased yields due to increased retention of pods and increased weight of seeds. El-Antably (1975), stated that the application of chlormequat to *Vicia faba* resulted in increases in the levels of endogenous IAA, BA, GA and cytokinins of 10, 14, 5 and 3 folds respectively.

Daminozide

The growth regulatory properties of daminozide were first reported by Riddell *et al.* (1962). It is transported in both the phloem and xylem within the plant and is not broken down, thus, the concentration of daminozide only declines due to new growth (Dicks, 1976). However, in the soil, the chemical is rapidly broken down by microbial action (Steffens, 1979). Its most important use was to prevent pre-harvest drop, promote firmness and quality and induce early cropping in closely spaced apple orchards (Luckwill, 1976). It has also been shown to produce compact and even chrysanthemums (Garrod, 1982) increase yields of peanuts (Steffens, 1979) and increase the yield of canning size potato tubers (Laycock, 1971).

Application of daminozide to *Vicia faba*, was found to inhibit stem elongation and delay senescence. Modification of intra-plant competition by enhancing reproductive growth at the expense of vegetative growth, resulted in increased numbers of seeds per node (Chapman and Sadjadi, 1981). Abou-Elleil and El-Wazeri (1978) found that the application of daminozide increased pod set from 16 to 22% however, due to later pod drop, seed yield was decreased by 13%. Increased pod set, without the associated pod drop, resulted in increased yields of up to 81% (Smith^{M.L.}, unpublished). Increased yields of up to 27% were also recorded after the application of 8 kg / ha of Alar '85 (Dekker and Neuval, 1983).

However, at this rate of application the increased yield was considered to be uneconomical.

Paclobutrazol

Paclobutrazol (PP333), a triazole growth retardant, which inhibits gibberellin production (Hedden and Graebe, 1985), was first synthesized by ICI^{plc.} in 1976. It was evaluated as a growth retardant to reduce lodging, and increase grain yield in perennial rye-grass (*Lolium perenne*), (Hebblethwaite *et al.* 1982) and is currently marketed to reduce vegetative growth and increase bud formation in top fruit (Lenton, 1987). PP333 has also been shown to reduce stem length in wheat by reducing basal internode length, leading to reduced lodging and increased yield (Frogatt *et al.*, 1982). Paclobutrazol is mainly a growth regulator, but as a triazole, also has some fungicide properties (Frogatt *et al.*, 1982; Lenton, 1987). PP333 is xylem mobile and so to give effective retardation its primary route of uptake is by the roots.

Application of PP333 to *Vicia faba* at the six leaf stage, resulted in shorter plants with increased harvest indices (Attiya *et al.*, 1983). Number of pods per plant was increased by 8% and seed yield per unit area was increased by 9%. Application of 1 kg/ha at the nine leaf stage, increased seed yield of Maris Bead by 42% and increased harvest index from 25.3% to 34.4% (Field *et al.*, 1989).

Flurprimidol

A formulation of flurprimidol (EL500), an experimental growth retardant from Dow Elanco, has been shown to have strong growth regulatory effects on winter oilseed rape; where reductions of up to 50% in stem height and reduced lodging have been recorded (Dawkins and Almond, 1984). Initial work on *Vicia faba* (Smyth, 1987), suggests that EL500 significantly increases pod set and yield.

Aims

The aim of the work presented here was to establish methods of increasing the yield of a commercial variety of broad bean by the use of plant growth substances. To this end, experimentation concentrated on three artificial plant growth retardants (EL500, JF 10405 and Alar), in an attempt to clarify the optimum application timing. Studies were also carried out to discover how levels of intrinsic hormones affected pod set. Using these findings in conjunction with findings from experiments that determined dry matter distribution throughout the growing season of treated plants, a more physiological approach to application timings could be made. Different methods of application of plant growth substances, fertilizer regimes and plant populations were also tested to assess whether these approaches may help to increase yield and stability of yield in a commercial situation.

Introduction

Both of the chapters relate to the experimental aspects of experimentation, however, this chapter refers to those techniques that were consistent throughout the whole of the thesis.

Material and Methods

Variety

In all experiments, the variety of broad bean used was Thorold White, supplied by Elmore Seeds, Spalding, Leics. This variety was chosen because of its widespread commercial usage.

Chemicals

The chemicals used are described in Table 2.1.

Table 2.1: Chemicals used in the experiments

Trade Name	Active Ingredient	Supplier
Agriol	ant-ionic water	ICI Agrochemicals
Alar	hexachloride	Dow Elanco Ltd
GA ₃	6-benzylaminopurine	SIGMA Chemical Co. Ltd
Banvol	benzoyl	De Ben (UK) Ltd
CLIAA	2-chlorobenzal	SIGMA Chemical Co. Ltd
EL 390	a formulation of terbufosfate	Dow Elanco Ltd
GA ₄	gibberellic acid	SIGMA Chemical Co. Ltd
Gibberox	gibberox	ICI Agrochemicals
NaOH	sodium hydroxide	SIGMA Chemical Co. Ltd
IP 10005	a formulation of peribenzox	ICI Agrochemicals

Introduction

Individual chapters relate to the actual procedure of experimentation, however, this chapter refers to those techniques that were consistent throughout the whole of the thesis.

Variety

In all experiments, the variety of Broad bean used was Threefold White, supplied by Elsoms Seeds, Spalding, Lincs. This variety was chosen because of its widespread commercial useage.

Chemicals

The chemicals used are described in Table 2.1.

Table 2.1: Chemicals used in the experiments

Trade Name	Active ingredient	Supplier
Agral	non-ionic wetter	ICI Agrochemicals plc
Alar	daminozide	Dow Elanco Ltd.
BAP	6-benzylaminopurine	SIGMA Chemical Co. Ltd.
Benlate	benomyl	Du Pont (UK) Ltd.
CLIAA	4-chloroindole	SIGMA Chemical Co. Ltd.
EL500	a formulation of flurprimidol	Dow Elanco Ltd.
GA ₃	gibberellic acid	SIGMA Chemical Co. Ltd.
Gramoxone	paraquat	ICI Agrochemicals plc
NaOH	sodium hydroxide	SIGMA Chemical Co. Ltd.
JF 10405	a formulation of paclobutrazol	ICI Agrochemicals plc

Enumeration of Flowers

Flower position within a raceme was assigned a number, 1 being the most proximal, 2 the next and so on (Fig. 2.1). Racemes were numbered acropetally, thus the lowest raceme was numbered 1, the next 2 and so on until the top of the plant was reached. In this way, each and every flower on the mainstem was assigned its own number. This allowed dates of tripping and chemical application to be recorded accurately, as well as allowing detailed analysis of flower and pod drop to be performed.

Growth Stages

A system of growth stages (Table 2.2), was developed to allow accurate identification of chemical application in different experiments. It also allowed accurate identification of growth stage within the text, without the excessive use of words.

The basis of the growth stages used in the thesis, was designed around an initial key developed by Smith (1982). This described the stages of flower development (F.D.), from bud initiation to full development (Fig. 2.2). The process of pod set was judged complete, when a young green pod of 1-2cm in length was clearly visible after petal abscission.

Hand-Tripping

When plants were grown in the bee-proof glasshouses, it was necessary to hand-trip the flowers in order to ensure that the flowers were pollinated. Hand-tripping is achieved by holding the standard petal and depressing the keel petal.

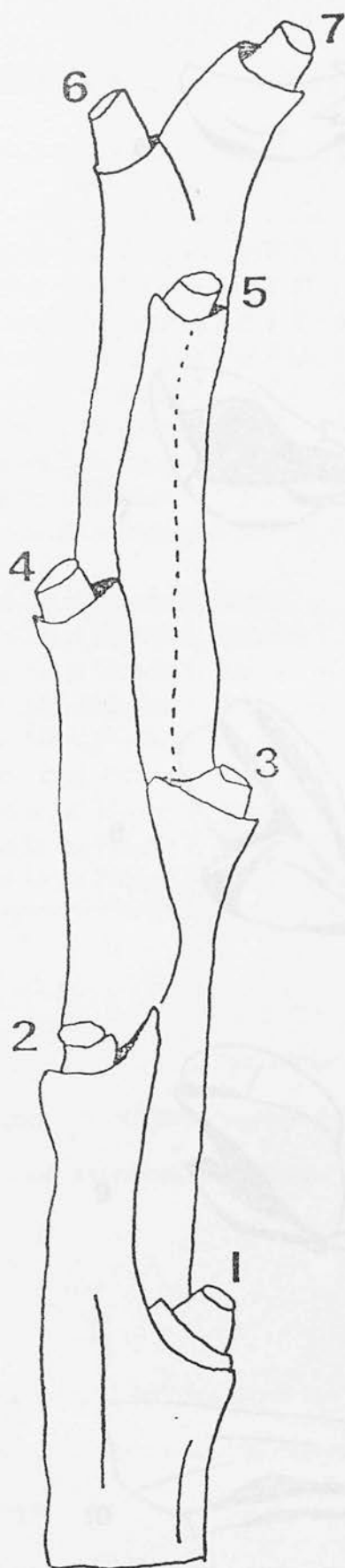


Figure 2.1: Enumeration of flowers within a raceme in *Vicia faba* (After Smith, 1982).

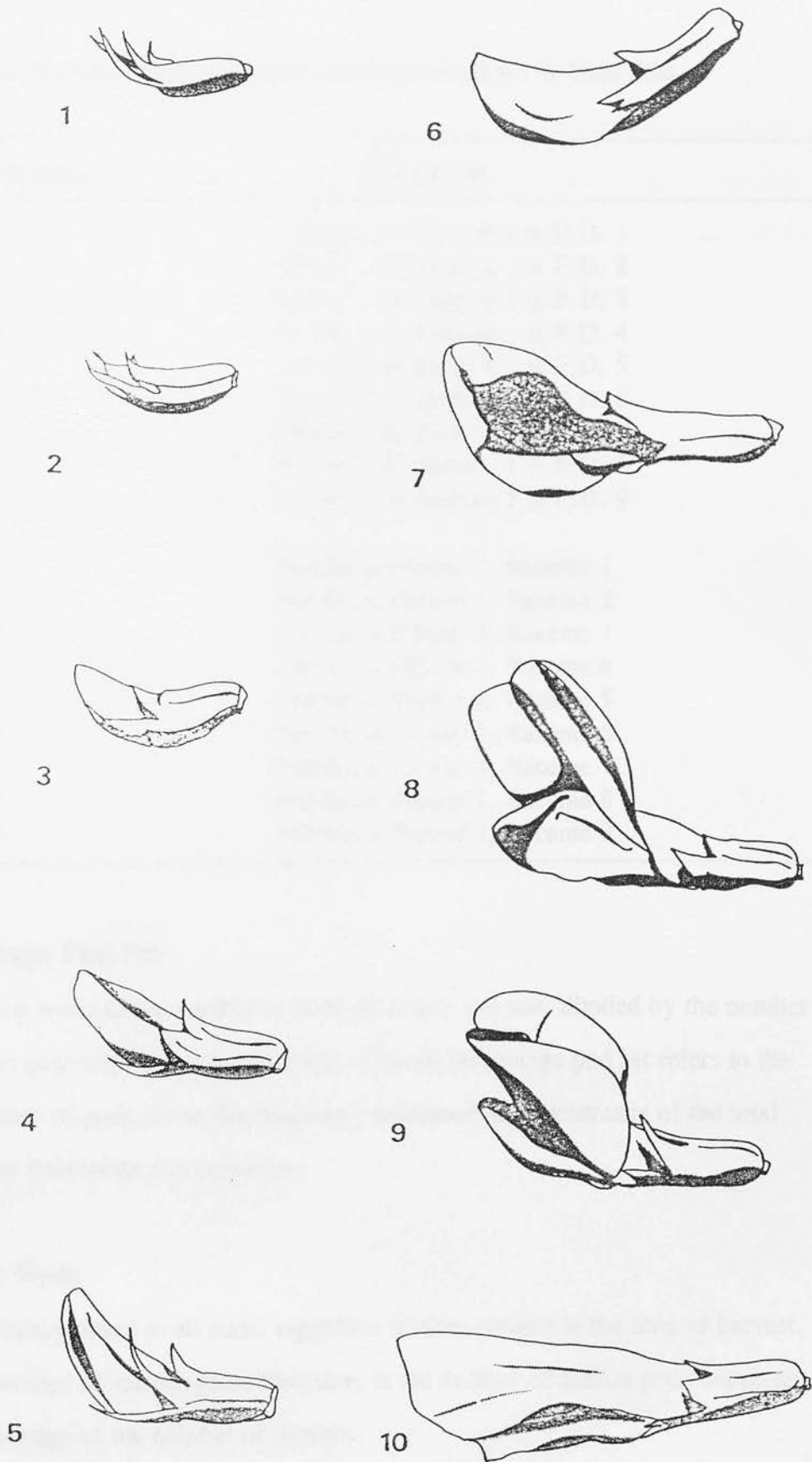


Figure 2.2: Stages of flower development in *Vicia faba* (After Smith, 1982)

Table 2.2: Growth Stages of Reproductive Development in *Vicia faba*.

Growth Stage	Description
01	Flower 1 on Raceme 1 at F.D. 1
02	Flower 1 on Raceme 1 at F.D. 2
03	Flower 1 on Raceme 1 at F.D. 3
04	Flower 1 on Raceme 1 at F.D. 4
05	Flower 1 on Raceme 1 at F.D. 5
06	Flower 1 on Raceme 1 at F.D. 6
07	Flower 1 on Raceme 1 at F.D. 7
08	Flower 1 on Raceme 1 at F.D. 8
09	Flower 1 on Raceme 1 at F.D. 9
11	Pod Set at Flower 1, Raceme 1
21	Pod Set at Flower 1, Raceme 2
31	Pod Set at Flower 1, Raceme 3
41	Pod Set at Flower 1, Raceme 4
51	Pod Set at Flower 1, Raceme 5
61	Pod Set at Flower 1, Raceme 6
71	Pod Set at Flower 1, Raceme 7
81	Pod Set at Flower 1, Raceme 8
91	Pod Set at Flower 1, Raceme 9

Percentage Pod Set

This figure refers to the number of pods set at any one site, divided by the number of flowers positions relevant to that site. Overall percentage pod set refers to the total number of pods set on the mainstem, expressed as a percentage of the total number of flowers on the mainstem.

Mature Pods

This definition refers to all pods, regardless of size, present at the time of harvest.

The percentage of mature pods, therefore, is the number of mature pods expressed as a percentage of the number of flowers.

Harvestable Pods

Harvestable pods are those pods that were of a harvestable size. Although somewhat arbitrary, this description was reserved for those pods that had attained a size that would support beans of a size suitable for eating. Percentage harvestable pods is defined as the number of harvestable pods, expressed as a percentage of the number of flowers.

Pod Retention

Percentage pod retention was defined as being the number of harvestable pods expressed as a percentage of the number of pods set.

Introduction

Experiments using plant growth regulators (Smith, 1982 ; Keller and Belluci, 1983), have shown that their exogenous application can significantly increase the pod set and yield of *Vicia faba*.

The use of daminozide to alter the growth characteristics of the plant in favour of reproductive growth has been studied by several workers. Chapman and Sadjadi (1981) applied daminozide to the cut surface of a decapitated plant, with the result that seed numbers per node increased by 21%. Abou-Elleil and El-Wazeri (1978) applied the chemical 1 and 3 months after planting and found that the use of daminozide increased flower retention. However, the final yield was reduced due to extra pod drop during the later stages of plant growth. Daminozide (85%) applied at 8 Kg/ha, at the beginning of flowering resulted in 10 and 27% yield increases during the 1980 and 1981 growing seasons respectively (Dekker and Neuval, 1983).

Paclobutrazol (PP333), was sprayed at the 6 leaf stage by Attiya *et al.* (1983). Treated plants were found to be shorter and had a greater yield / m², due to a slight increase (8%), in the number of pods per plant. Application of paclobutrazol at the 2 leaf stage in combination with naphthalene-acetic acid (NAA) and benzyladenine (BA) at flowering to increase sink strength caused a significant increase in the number of pods at lower racemes. This yield advantage was again lost due to later pod drop.

A formulation of flurprimidol (EL500), has been shown to exhibit strong regulatory effects on *Vicia faba* grown in the glasshouse, causing decreased flower drop and increased numbers of mature pods (Smyth, 1987).

The aim of this experiment was to determine how these three plant growth regulators applied at specific growth stages affected the growth characteristics of the broad bean variety Threefold White. Application timings were based upon flower opening (as described in Chapter 2). Exact time of application could therefore be determined and so compared to future experiments.

Method

Seeds of the variety Threefold White were sown on at the Bush Estate, Edinburgh School of Agriculture on 22 April 1987. The experiment was designed as a completely randomised layout, each treatment (Table 3.1), contained 3 replicated plots. Each plot measured 1 x 1.5m and seeds were sown at regular intervals along 30 cm rows in order to attain a plant population of 30 plants m⁻². A distance of 1m was left between each plot.

The previous crop was barley. Soil samples indicated that the indices for P and K were 3 in both cases, so it was considered unnecessary to place any compound fertilizer in the seed bed, no nitrogen was applied. No herbicides were used on the plots, However, during the early stages of growth the plots were kept weed free by hoeing. Chocolate spot was treated at early pod set (July 23) with Benlate (benomyl), at the rate of 1 Kg/ha.

The growth regulators used were Alar '85 (daminozide), at a rate of 1g/litre, JF 10405 (a formulation of paclobutrazol), at a rate of 2ml/litre and EL500 (flurprimidol) at a rate of 0.25g/litre. All applications were made with a Cooper Pegler knapsack sprayer fitted with fan-jet nozzles and were applied to "run off". This represented an application of 1500 Litres/ha. Timing of applications is described in Table 3.1. All treatments contained the non-ionic wetter "Agral", at a

rate of 1 ml/litre. Control plants were sprayed with a mixture of water containing the wetter at growth stage 09.

Table 3.1: Treatments applied during the experiment.

Treatment	Timing of application (Growth Stage)			
	01	03	07	09
Control				Water + Wetter
A	EL500			
B	JF 10405			
C	ALAR			
D		EL500		
E		JF 10405		
F		ALAR		
G			EL500	
H			JF 10405	
I			ALAR	
J				EL500
K				JF 10405
L				ALAR

In order to prevent edge effects, 5 plants were chosen at random from within the centre of the plots and tagged to allow accurate identification. Plants were scored for number of flowers formed, pod set and pod drop. At harvest (14 - 15 September), the same plants were scored for vegetative (distance from soil surface to the first flowering raceme) and reproductive height (distance from the first flowering raceme to the apex of the plant). Yield analysis was split into number of mature pods (those retained by the plant until harvest), number of harvestable pods (those pods considered to be of a harvestable size), weight of harvestable pods, number of seeds and weight of seeds on a whole plant basis.

Results

Intra-Raceme Pod Set

Control Plants

The pattern of pod set on control plants demonstrated a typical pattern, with most pods setting at proximal positions within the raceme. The first three flowers formed had an average percentage pod set of 31.6%, while the 3 distal flower positions had an average percentage set of 0.9%.

Effect of EL500

EL500 applied at growth stage 01 (Treatment A), caused increased pod set at all flower positions compared to control plants. This increase in pod set was significant ($p < 0.05$), at flower position 2 and caused the average pod set over the first three flowers to increase to 43.2% (Fig. 3.1a). Pod set at the distal 3 flowers averaged 5.2%.

At growth stage 03 pod set was 26.3% ($p < 0.001$) and 23.6% ($p < 0.01$) greater at flower positions 2 and 3 respectively, compared to control plants (Fig. 3.1b).

Generally more pods set compared to when the application was made at growth stage 01, but this effect was not significant.

Application of EL500 at growth stage 07 (Treatment G), resulted in 22.3%, 34.2% and 30.6% ($p < 0.001$) more pod set compared to control plants at flowers 2, 3 and 4 (Fig. 3.1c). Generally, increases in pod set were evident over Treatment D.

Increases in pod set were evident at all flower positions compared to Treatment A, with significant increases of 21.4% and 18.8% ($p < 0.01$) evident at flowers 3 and 4.

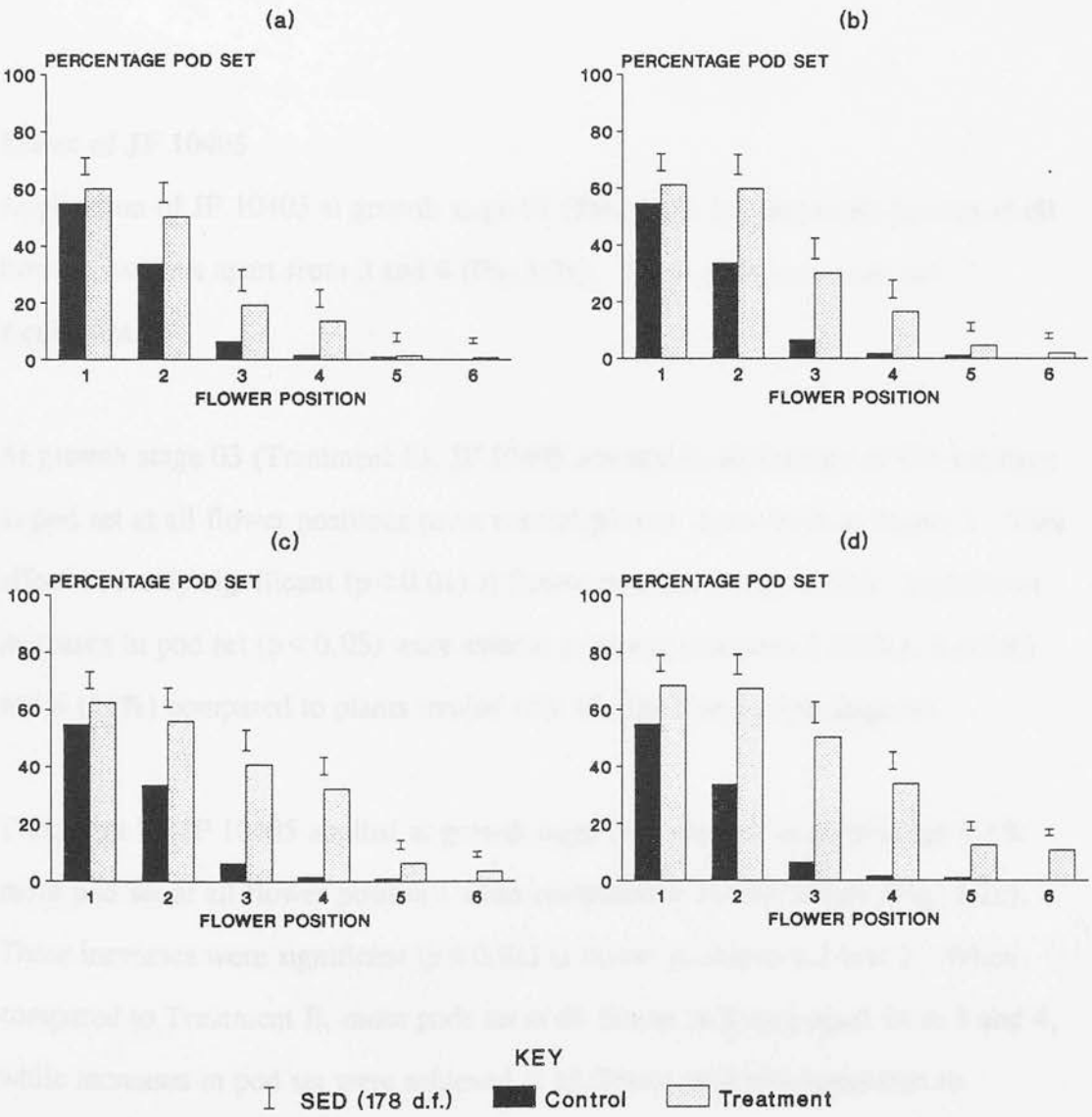


Figure 3.1: Effect of EL500 applied at a) growth stage 01, b) growth stage 03, c) growth stage 07 and d) growth stage 09, on intra-raceme percentage pod set. (Actual figures are shown in Appendix 3.1)

At growth stage 09, pod set was increased by on average 24% ($p < 0.01$), at all flower positions compared to control plants (Fig. 3.1d). In general, increased pod set at all flower positions was also evident compared to all other EL500 treatments regardless of timing.

Effect of JF 10405

Application of JF 10405 at growth stage 01 (Treatment B), decreased pod set at all flower positions apart from 3 and 4 (Fig.3.2a). These decreases were not significant.

At growth stage 03 (Treatment E), JF 10405 resulted in an average of 8% increase in pod set at all flower positions (over control plants), apart from at flower 1. This effect was only significant ($p < 0.01$) at flower position 3 (Fig. 3.2b). Significant increases in pod set ($p < 0.05$) were evident at flower positions 2 (15%), 3 (19%) and 4 (11%) compared to plants treated with JF 10405 at growth stage 01.

Treatment H (JF 10405 applied at growth stage 07), resulted in an average 9.1% more pod set at all flower positions when compared to control plants (Fig. 3.2c). These increases were significant ($p < 0.01$) at flower positions 1,2 and 3. When compared to Treatment E, more pods set at all flower positions apart from 3 and 4, while increases in pod set were achieved at all flower positions compared to Treatment B.

Treatment K (JF 10405 applied at growth stage 09), caused 18.9% more pods to set at the first four flower positions ($p < 0.001$) compared to control plants (Fig. 3.2d).

Figure 3.2: Effect of JF 10405 applied at a) growth stage 01, b) growth stage 03, c) growth stage 07, and d) growth stage 09, on total relative percentage pod set. (Actual figures are shown in Appendix 3.1)

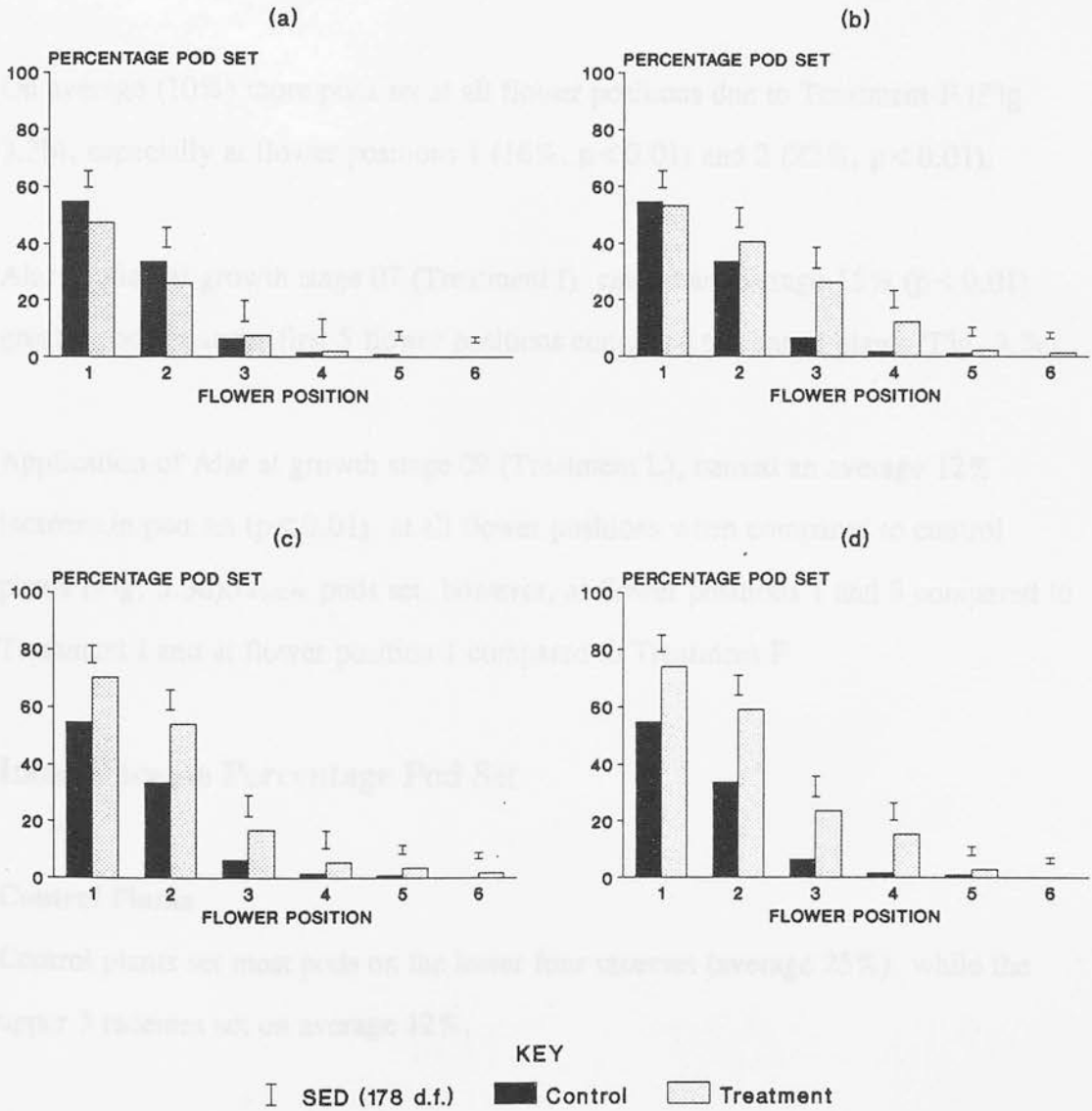


Figure 3.2: Effect of JF 10405 applied at a) growth stage 01, b) growth stage 03, c) growth stage 07 and d) growth stage 09, on intra-raceme percentage pod set. (Actual figures are shown in Appendix 3.1)

Effect of Alar

Treatment C (Alar applied at growth stage 01), resulted in increases in pod set at the first 5 flower positions compared to control plants (Fig 3.3a). Significant increases ($p < 0.01$) were achieved at flower positions 2 (19%) and 3 (15%).

On average (10%) more pods set at all flower positions due to Treatment F (Fig 3.3b), especially at flower positions 1 (16%, $p < 0.01$) and 2 (22%, $p < 0.01$).

Alar applied at growth stage 07 (Treatment I) caused an average 15% ($p < 0.01$) greater pod set at the first 5 flower positions compared to control plants (Fig. 3.3c).

Application of Alar at growth stage 09 (Treatment L), caused an average 12% increase in pod set ($p < 0.01$) at all flower positions when compared to control plants (Fig. 3.3d). FEWER pods set however, at flower positions 1 and 3 compared to Treatment I and at flower position 1 compared to Treatment F.

Inter-Raceme Percentage Pod Set

Control Plants

Control plants set most pods on the lower four racemes (average 25%) while the upper 3 racemes set on average 12%.

Effect of EL500

Treatment A resulted in an average 12% increase in pod set at all racemes. Increases on the first four racemes were not significant, however, on the upper 3 racemes average pod set was increased to 27% ($p < 0.01$, Fig. 3.4a).

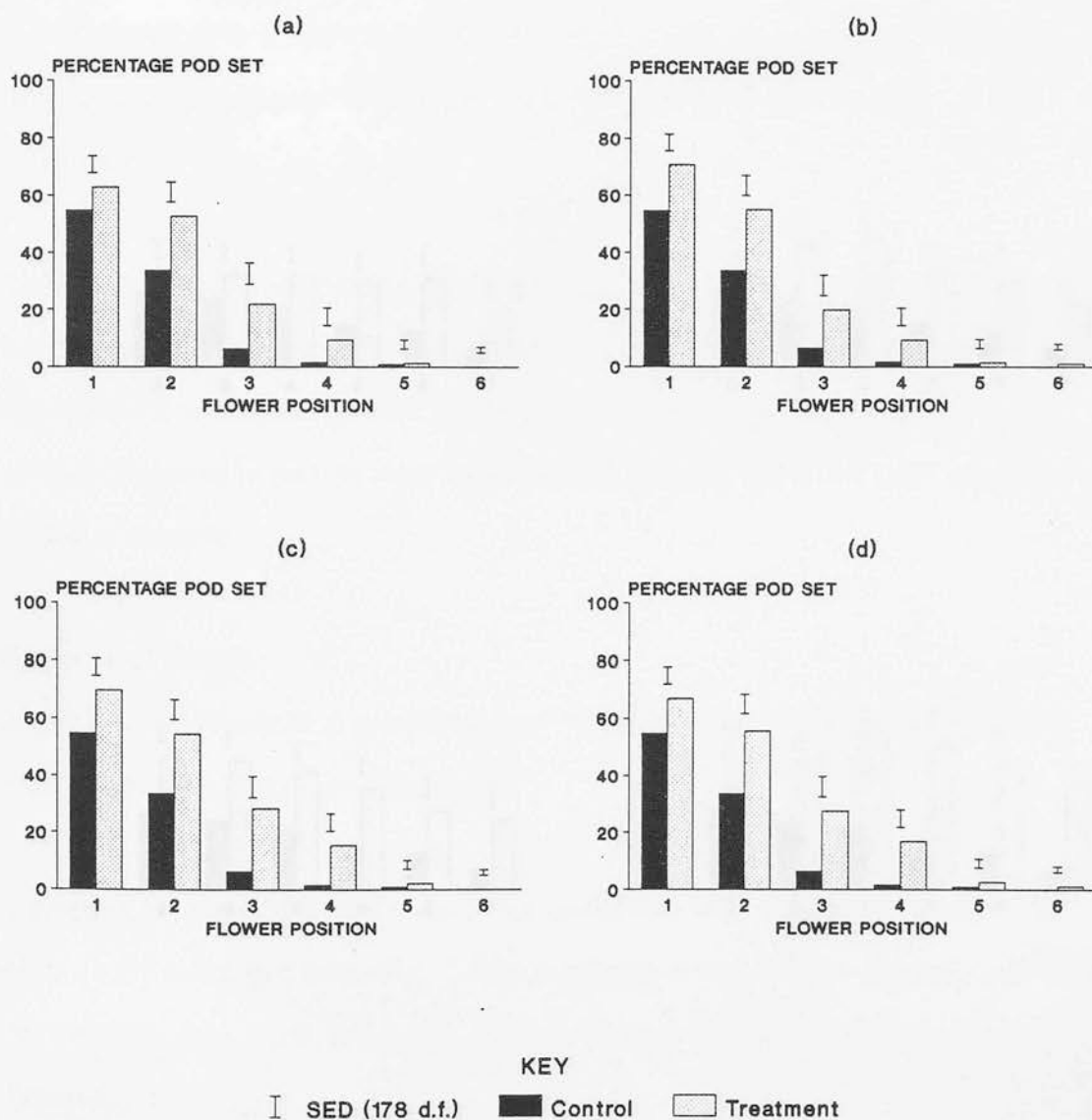


Figure 3.3: Effect of Alar applied at a) growth stage 01, b) growth stage 03, c) growth stage 07 and d) growth stage 09, on intra-raceme percentage pod set. (Actual figures are shown in Appendix 3.1)

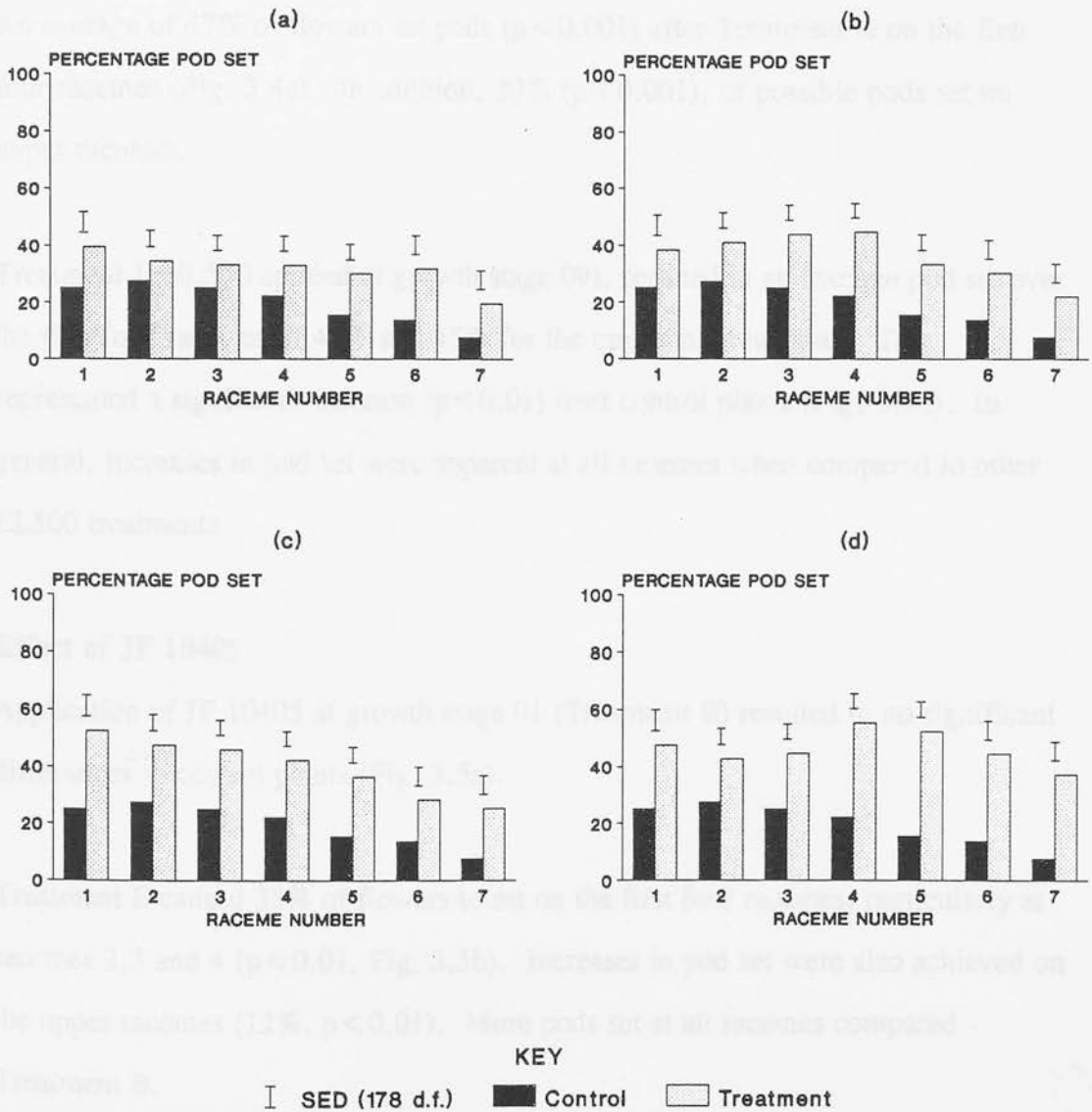


Figure 3.4: Effect of EL500 applied at a) growth stage 01, b) growth stage 03, c) growth stage 07 and d) growth stage 09, on inter-raceme percentage pod set. (Actual figures are shown in Appendix 3.2)

More pods set after the application of EL500 at growth stage 03, when compared to control plants. Average pod set on the lower four racemes was 42% ($p < 0.01$), while on the upper 3 racemes it was 29% ($p < 0.001$, Fig. 3.4b).

An average of 47% of flowers set pods ($p < 0.001$) after Treatment G on the first four racemes (Fig. 3.4c). In addition, 30% ($p < 0.001$), of possible pods set on upper racemes.

Treatment J (EL500 applied at growth stage 09), resulted in an average pod set over the first four racemes of 48% and 45% for the upper three racemes. This represented a significant increase ($p < 0.01$) over control plants (Fig. 3.4d). In general, increases in pod set were apparent at all racemes when compared to other EL500 treatments.

Effect of JF 10405

Application of JF 10405 at growth stage 01 (Treatment B) resulted in no significant differences ^{OVER} control plants (Fig. 3.5a).

Treatment E caused 35% of flowers to set on the first four racemes, particularly at racemes 2,3 and 4 ($p < 0.01$, Fig. 3.5b). Increases in pod set were also achieved on the upper racemes (12%, $p < 0.01$). More pods set at all racemes compared Treatment B.

Treatment H resulted in an average of 33% pod set on the first four racemes and 27% on the upper racemes (Fig 3.5c). On average, 13% more flowers set pods on each raceme compared to treatment B. FEWER pods set on racemes 3,4 and 5, when compared to Treatment E, however, this decrease was only significant ($p < 0.05$) at raceme 3 (11%).

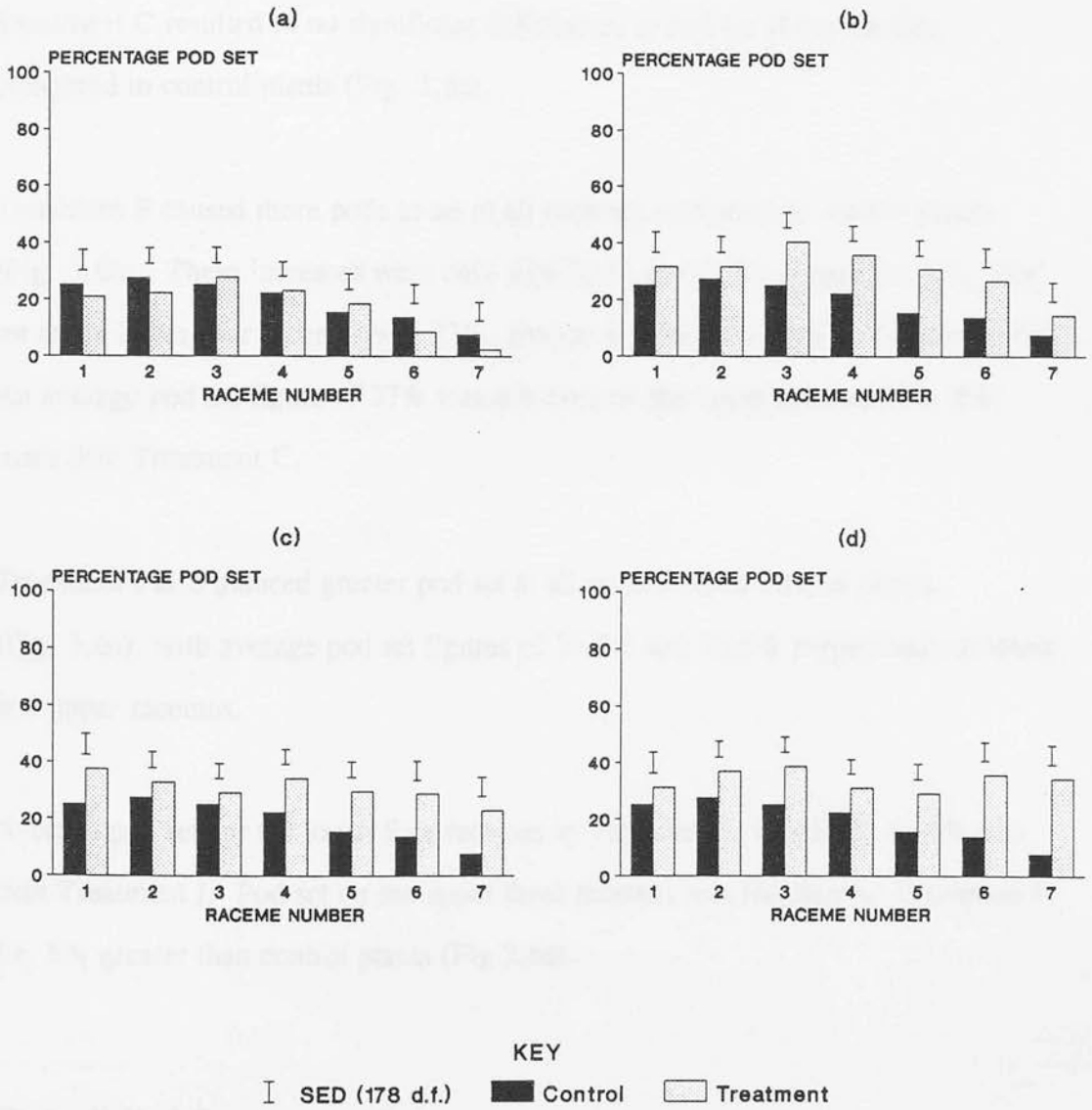


Figure 3.5: Effect of JF 10405 applied at a) growth stage 01, b) growth stage 03, c) growth stage 07 and d) growth stage 09, on inter-raceme percentage pod set. (Actual figures are shown in Appendix 3.2)

Treatment K increased pod set on the first four racemes to an average of 34%, with pod set also increased to 33% on the upper 3 racemes (Fig. 3.5d).

Effect of Alar

Treatment C resulted in no significant differences in pod set at any raceme compared to control plants (Fig. 3.6a).

Treatment F caused more pods to set at all racemes compared to control plants (Fig. 3.6b). These increases were only significant ($p < 0.05$) at racemes 4-7. Pod set at the lower four racemes was 32%, similar to levels achieved by Treatment C. An average pod set figure of 27% was achieved on the upper racemes, i.e. 6% more than Treatment C.

Treatment I also induced greater pod set at all racemes over control plants (Fig. 3.6c) with average pod set figures of 36.3% and 25.9% respectively at lower and upper racemes.

Average pod set for the lower four racemes in Treatment L was 34%, or 2% less than Treatment I. Pod set on the upper three racemes was identical to Treatment I, i.e. 6% greater than control plants (Fig 3.6d).

Overall Pod Set

Overall pod set increased significantly ($p < 0.05$), due to application of all plant growth regulators, at all timings apart from Treatment E, where there was no difference and at Treatment B, where a decrease of 2.4% occurred (Table 3.2). In general, the later the timing, the greater the increase in percentage pod set compared to control plants (Fig. 3.7)

Table 3.2: The effect of EL500, JF 10405 and Alar applied at four separate timings on number of flowers, pod set, mature pods and harvestable pods.

Treatment	No. Flowers	No. Pods Set	% Pods Set	No. Mature Pods	% Mature Pods	No. Harvest Pods	% Harvest Pods
Control	50.9	8.5	17.2	4.6	9.6	4.3	8.9
A	65.5	15.5	24.2	7.9	12.2	7.3	11.3
B	56.7	8.1	14.8	4.2	7.6	4.1	7.4
C	53.9	13.8	24.8	6.1	11.1	5.5	10.0
D	56.5	16.6	30.0	8.0	14.3	6.5	11.7
E	65.2	14.5	23.3	6.9	10.8	6.5	10.2
F	56.3	14.8	26.8	5.7	10.0	5.5	9.8
G	58.1	19.1	32.9	7.5	13.0	6.3	10.7
H	51.7	13.9	27.3	5.9	11.8	5.6	11.2
I	49.4	14.7	29.8	6.3	12.8	5.9	12.0
J	52.7	21.6	42.0	8.7	16.7	7.3	14.1
K	49.0	15.9	33.3	6.1	13.0	5.5	11.6
L	54.5	15.5	28.7	7.3	13.4	6.7	12.3
SED(178 df)	4.55	1.93	3.23	0.90	1.54	0.81	1.42
f value	0.003	<0.001	<0.001	<0.001	<0.001	<0.001	0.002

All figures represent the mean of 5 plants

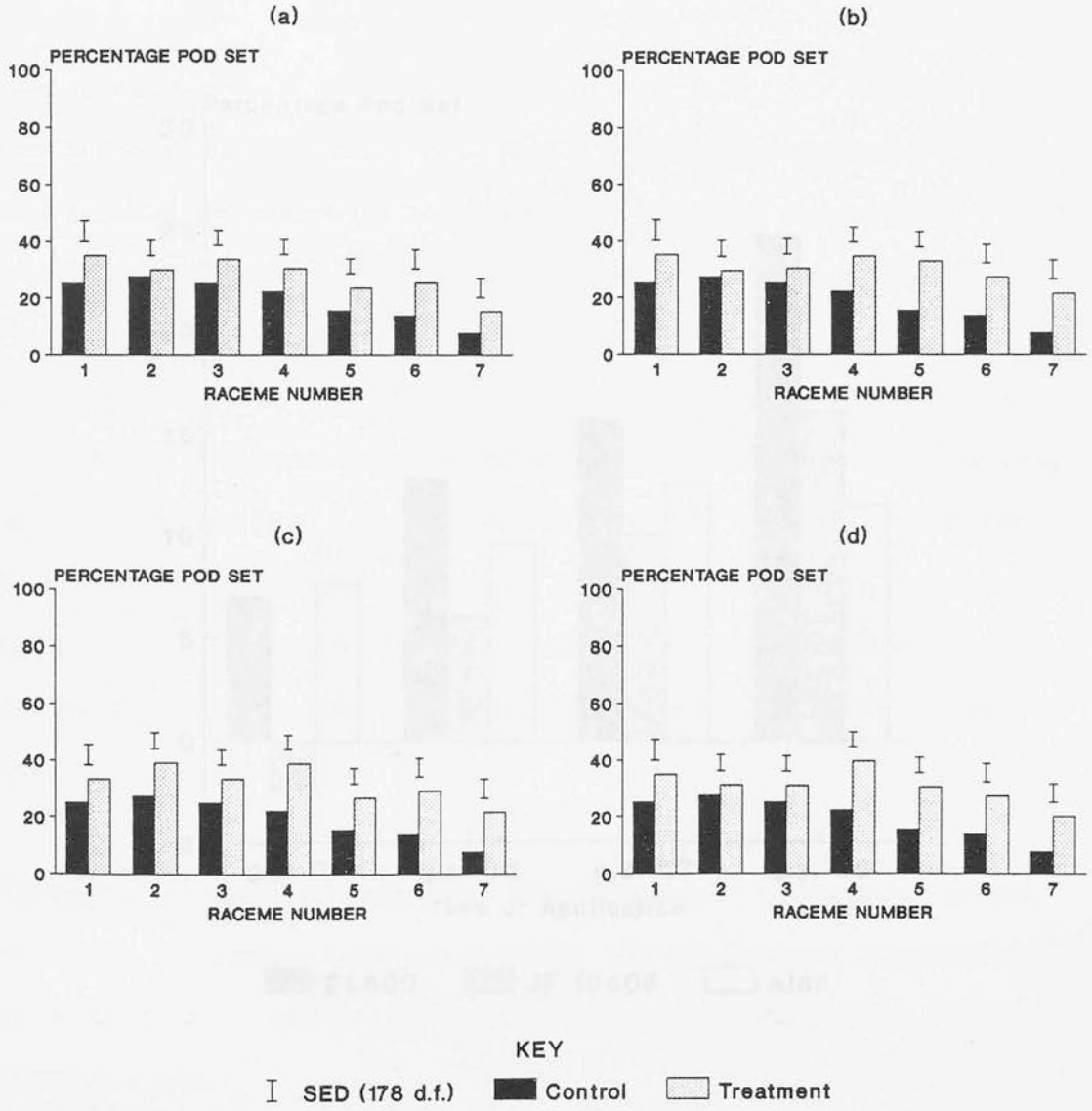


Figure 3.6: Effect of Alar applied at a) growth stage 01, b) growth stage 03, c) growth stage 07 and d) growth stage 09, on inter-raceme percentage pod set. (Actual figures are shown in Appendix 3.2)

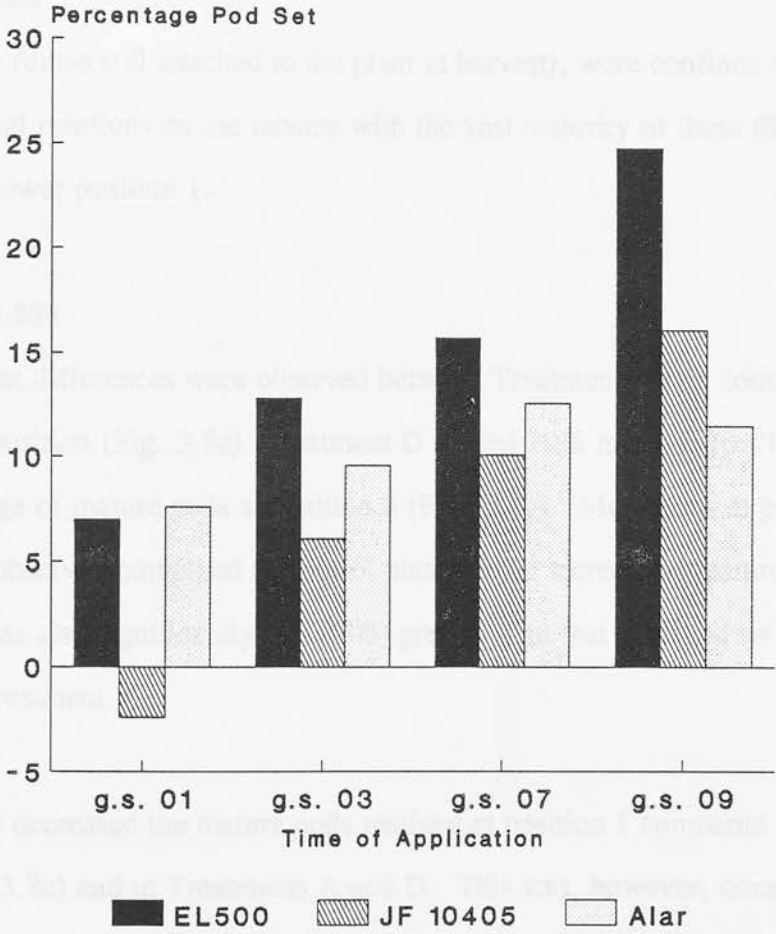


Figure 3.7: Effect of EL500, JF 10405 and Alar, applied at four separate growth stages on differences in overall percentage pod set, compared to control plants.

Only Alar deviated from this trend in that Treatment I had a greater percentage pod set than Treatment L.

Intra-Raceme Percentage Mature Pods

Control Plants

Mature pods (those still attached to the plant at harvest), were confined to the two most proximal positions on the raceme with the vast majority of these (83%) situated at flower position 1.

Effect of EL500

No significant differences were observed between Treatment A and control plants at any flower position (Fig. 3.8a) Treatment D caused 20% increase ($p < 0.001$) in the percentage of mature pods at position 2 (Fig. 3.8b). More pods at positions 1, 3 and 4 were observed compared to control plants. The increase in mature pods at position 2 was also significantly ($p < 0.05$) greater than that achieved by plants subject to Treatment A.

Treatment G decreased the mature pods retained at position 1 compared to control plants (Fig. 3.8c) and to Treatments A and D. This was, however, compensated for by increased mature pods ($p < 0.001$), at positions 2 (18%) and 4 (14%), compared to control plants.

The application of EL500 at growth stage 09 (Treatment J), resulted in mature pods present at flower positions 1-5, with significant ($p < 0.01$), increases over control plants evident at positions 2, 3 and 4 (Fig. 3.8d). There were more pods at flower positions 4 and 5 compared to other EL500 treatments, though pod retention at other flower sites was similar to other treatments.

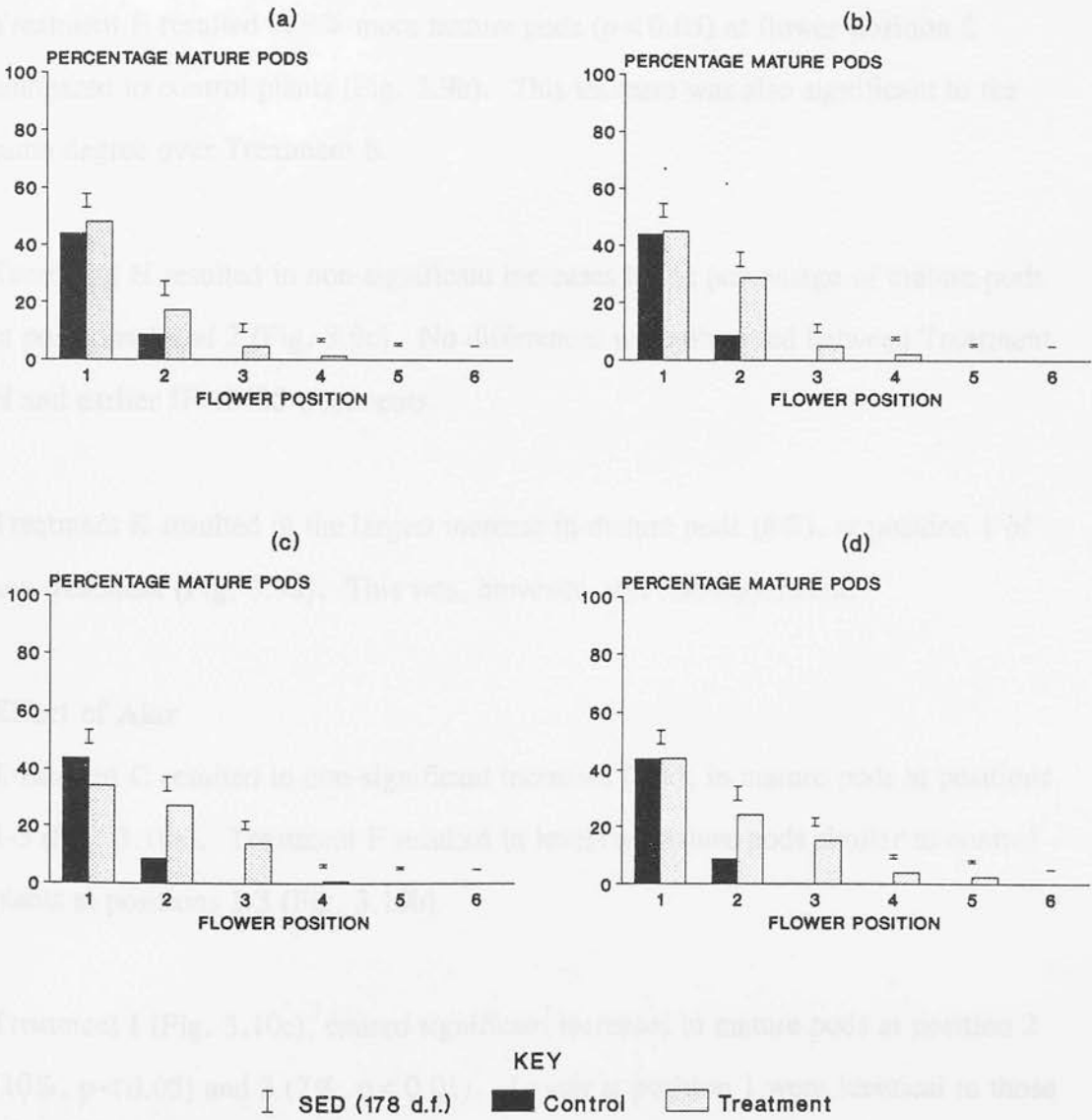


Figure 3.8: Effect of EL500 applied at a) growth stage 01, b) growth stage 03, c) growth stage 07 and d) growth stage 09, on intra-raceme percentage mature pods. (Actual figures are shown in Appendix 3.3)

Effect of JF 10405

Treatment B reduced by 10% ($p < 0.05$), mature pods at flower position 1 (Fig. 3.9a). No difference was observed at other flower positions.

Treatment E resulted in 9% more mature pods ($p < 0.05$) at flower position 2 compared to control plants (Fig. 3.9b). This increase was also significant to the same degree over Treatment B.

Treatment H resulted in non-significant increases in the percentage of mature pods at positions 1 and 2 (Fig. 3.9c). No differences were observed between Treatment H and earlier JF 10405 treatments.

Treatment K resulted in the largest increase in mature pods (8%), at position 1 of any treatment (Fig. 3.9d). This was, however, still non-significant.

Effect of Alar

Treatment C resulted in non-significant increases (3%), in mature pods at positions 1-3 (Fig. 3.10a). Treatment F resulted in levels of mature pods similar to control plants at positions 1-3 (Fig. 3.10b).

Treatment I (Fig. 3.10c), caused significant increases in mature pods at position 2 (10%, $p < 0.05$) and 3 (7%, $p < 0.01$). Levels at position 1 were identical to those of control plants. Increased levels of mature pods at positions 2 and 3 were also evident over earlier Alar treatments.

Treatment L significantly increased ($p < 0.05$), mature pods at positions 2 (16%) and 3 (6%), with increases also evident at positions 1 (4%) and 5 (1%), compared to control plants (Fig. 3.10d). More mature pods were retained at flower positions 1-3 than on plants subjected to the other Alar treatments.

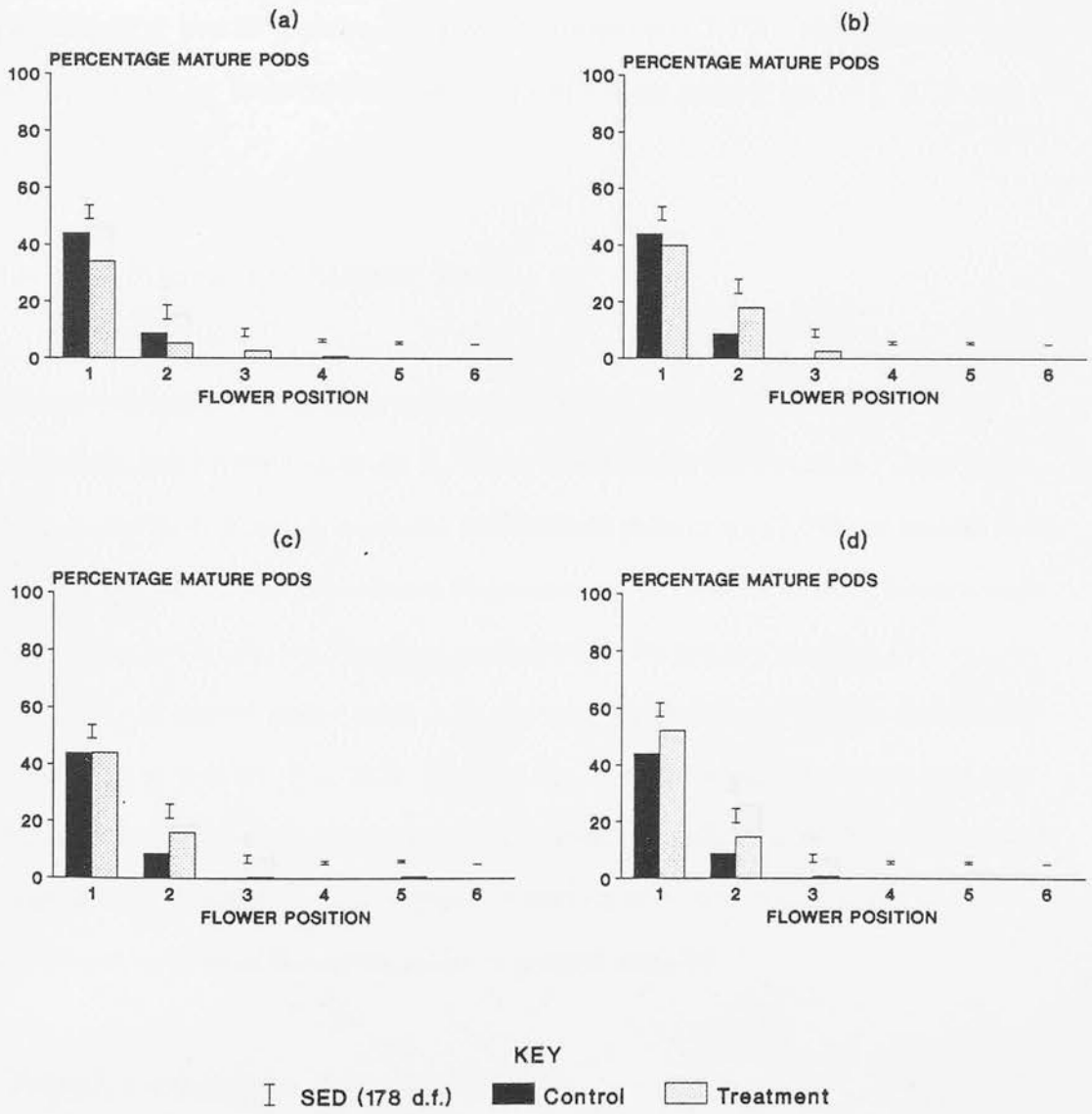


Figure 3.9: Effect of JF 10405 applied at a) growth stage 01, b) growth stage 03, c) growth stage 07 and d) growth stage 09, on intra-raceme percentage mature pods. (Actual figures are shown in Appendix 3.3)

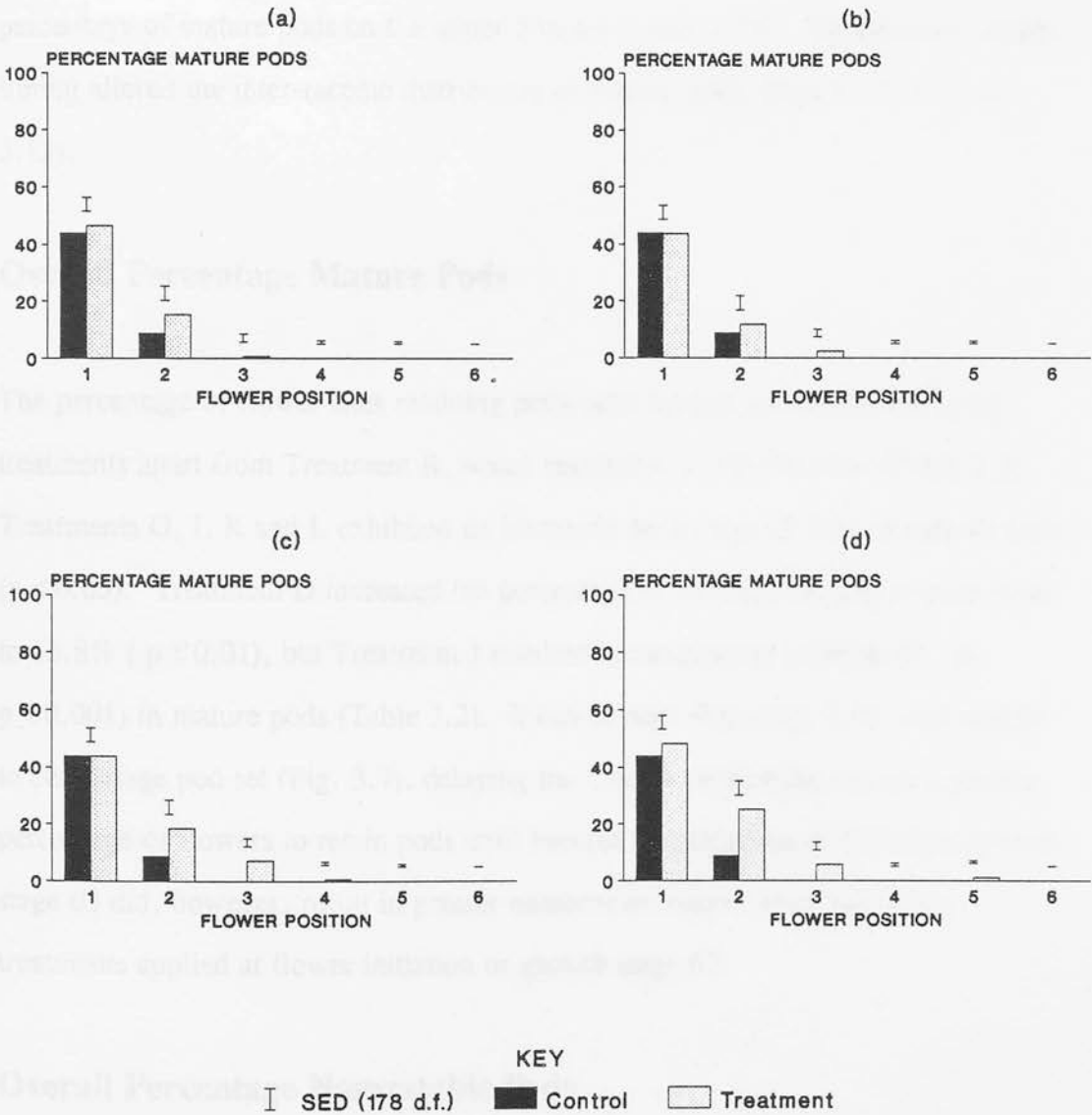


Figure 3.10: Effect of Alar applied at a) growth stage 01, b) growth stage 03, c) growth stage 07 and d) growth stage 09, on intra-raceme percentage mature pods. (Actual figures are shown in Appendix 3.3)

Inter-Raceme Percentage Mature Pods

On control plants, the majority of mature pods were confined to the lower four racemes, where the average percentage of mature pods was 16.5%, whereas average percentage of mature pods on the upper 3 racemes was 3.7%. No treatment at any timing altered the inter-raceme distribution of mature pods (Figs 3.11, 3.12 and 3.13).

Overall Percentage Mature Pods

The percentage of flower sites retaining pods until harvest was increased by all treatments apart from Treatment B, which resulted in a 2% decrease (Table 3.2). Treatments G, I, K and L exhibited an increased percentage (3.5%) of mature pods ($p < 0.05$). Treatment D increased the percentage of flowers forming mature pods to 13.8% ($p < 0.01$), but Treatment J resulted in the greatest increase (7.1%, $p < 0.001$) in mature pods (Table 3.2). It can be seen from Fig. 3.14 that similar to percentage pod set (Fig. 3.7), delaying the time of application caused a greater percentage of flowers to retain pods until harvest. Application of EL500 at growth stage 03 did, however, result in greater numbers of mature pods than either treatments applied at flower initiation or growth stage 07.

Overall Percentage Harvestable Pods

As with pod set and mature pods, the percentage of harvestable pods (those deemed to be of a size suitable for harvesting), followed a similar pattern (Fig. 3.15). Thus, in general, as the timing became later, the number of pods retained by the plant that developed into a harvestable size became greater. Again, the only exception to this rule was the application of EL500 at growth stage 03, which

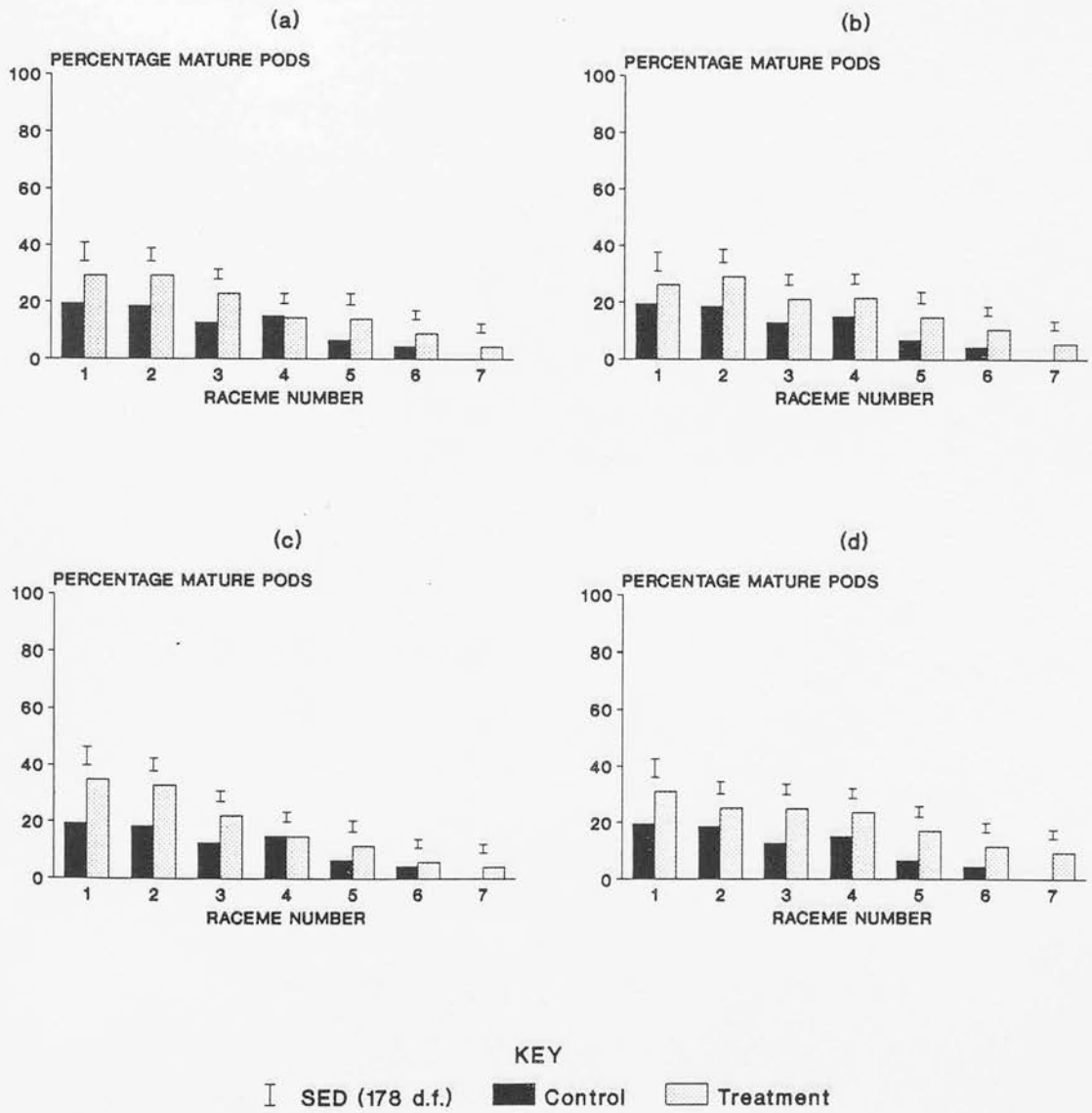


Figure 3.11: Effect of EL500 applied at a) growth stage 01, b) growth stage 03, c) growth stage 07 and d) growth stage 09, on inter-raceme percentage mature pods. (Actual figures are shown in Appendix 3.4)

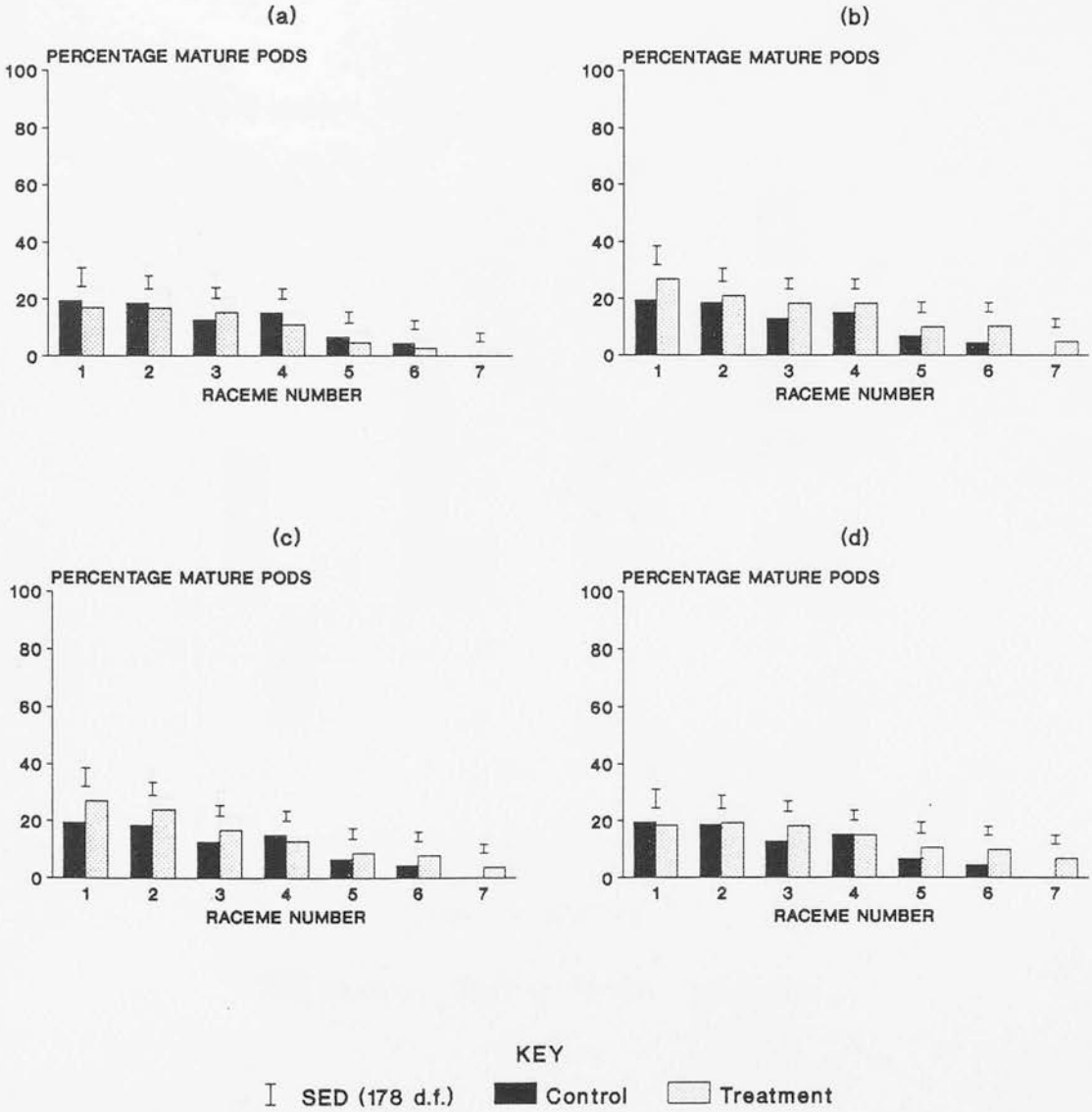


Figure 3.12: Effect of JF 10405 applied at a) growth stage 01, b) growth stage 03, c) growth stage 07 and d) growth stage 09, on inter-raceme percentage mature pods. (Actual figures are shown in Appendix 3.4)

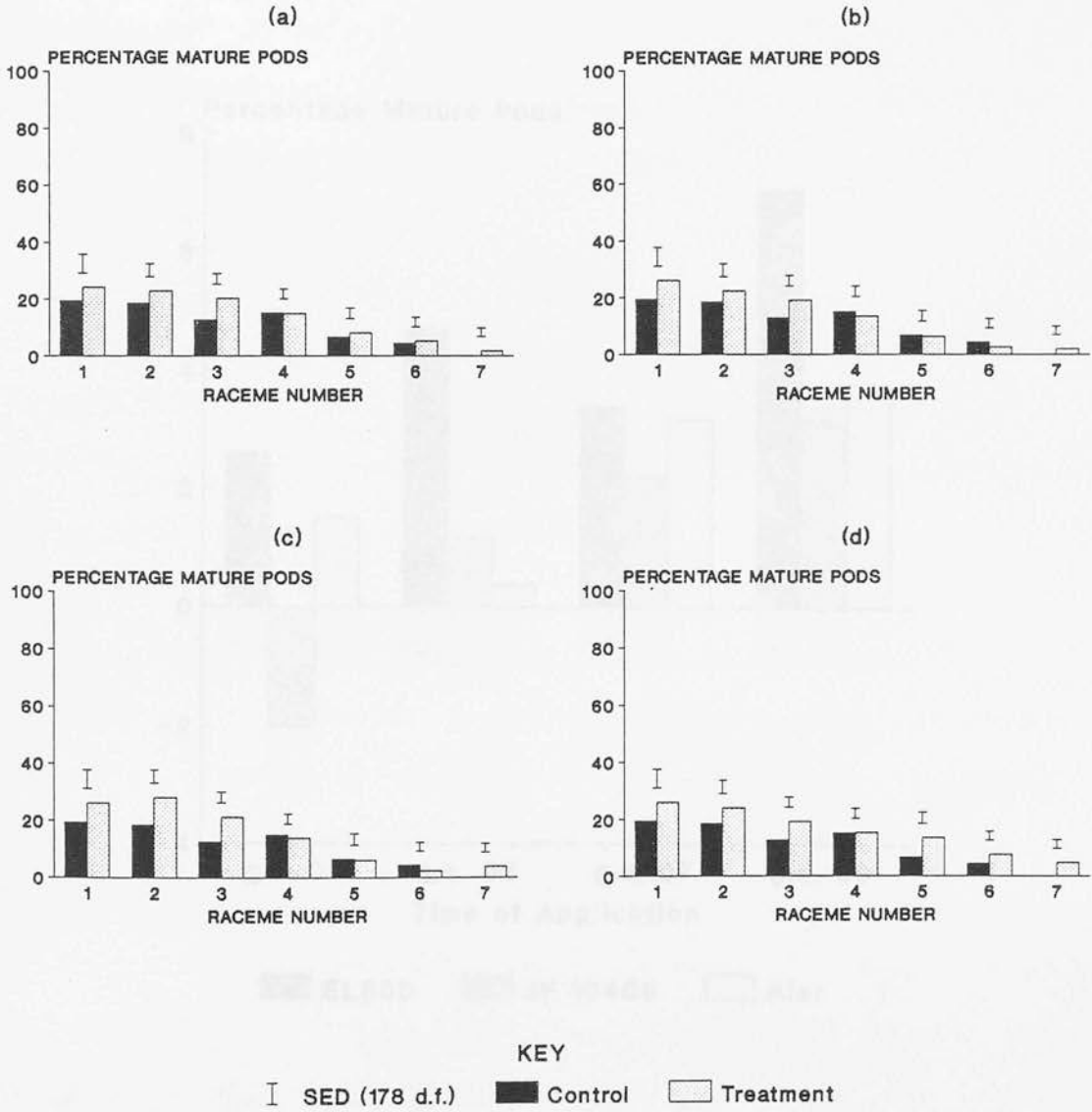


Figure 3.13: Effect of Alar applied at a) growth stage 01, b) growth stage 03, c) growth stage 07 and d) growth stage 09, on inter-raceme percentage mature pods. (Actual figures are shown in Appendix 3.4)

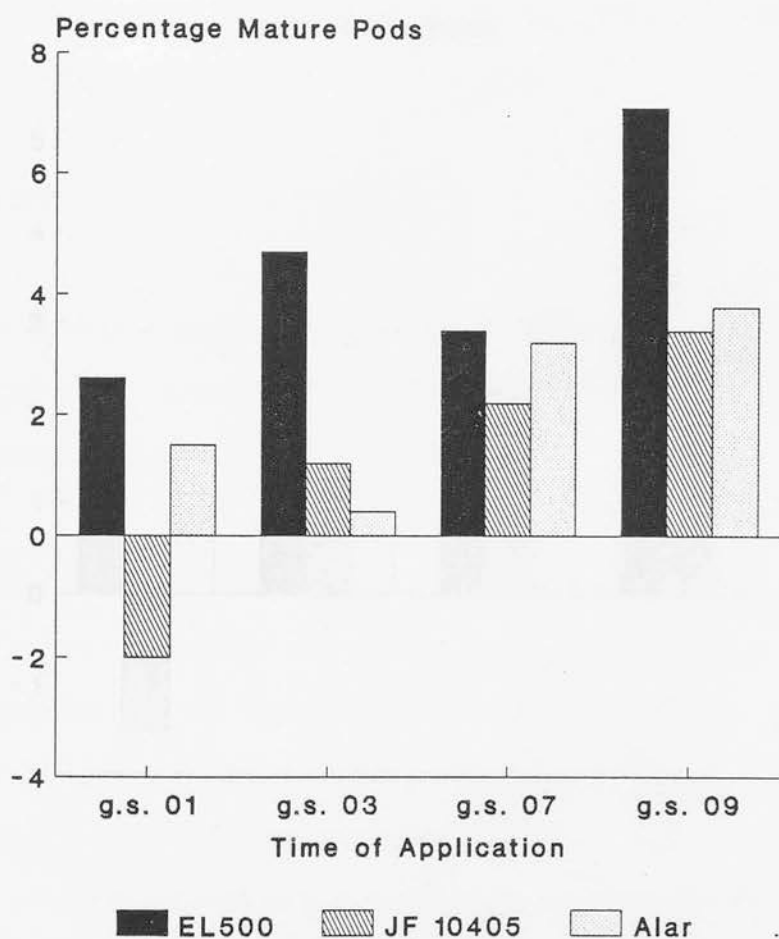


Figure 3.14: Effect of EL500, JF 10405 and Alar, applied at four separate growth stages on differences in overall percentage mature pods, compared to control plants.

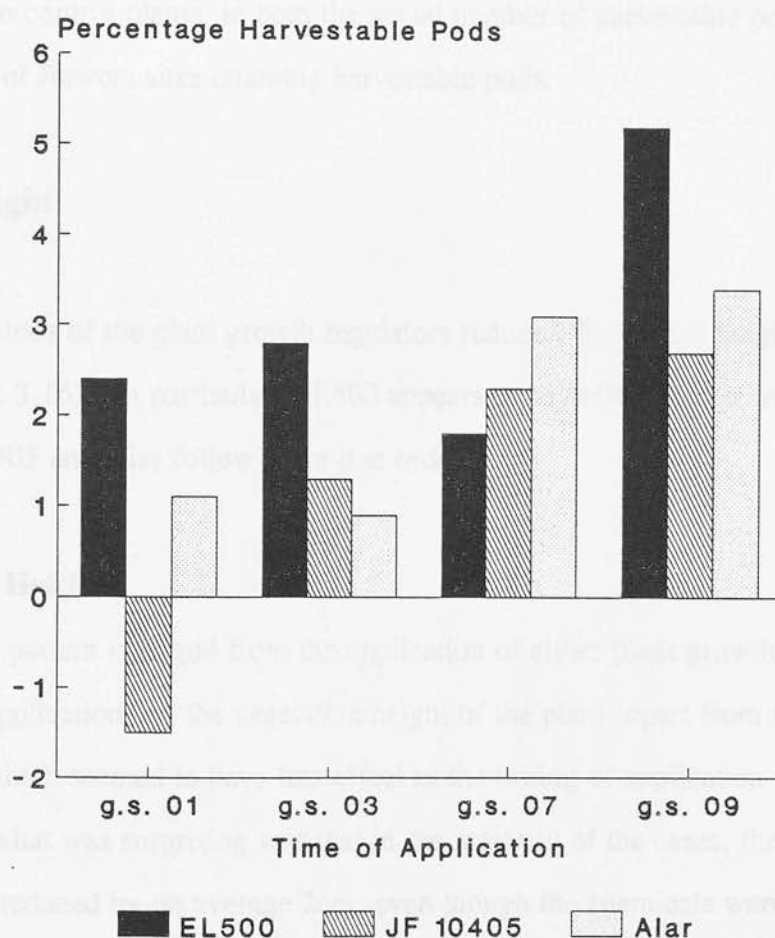


Figure 3.15: Effect of EL500, JF 10405 and Alar applied at four separate growth stages on the difference in overall percentage of harvestable pods, compared to control plants.

resulted in increased numbers of harvestable pods compared to flower initiation and growth stage 07 applications.

Treatment B remained the only treatment that caused a decrease in percentage harvestable pods. Treatment J resulted in a significant ($p < 0.001$) increase compared to control plants, in both the actual number of harvestable pods and the percentage of flowers sites retaining harvestable pods.

Plant Height

All applications of the plant growth regulators reduced the overall height of the plants (Fig. 3.16). In particular, EL500 appears to have the greatest overall effect with JF 10405 and Alar following in that order.

Vegetative Height

No definite pattern emerged from the application of either plant growth regulator or timing of application, on the vegetative height of the plant, apart from the effect of JF 10405 which seemed to have less effect as the timing of application was delayed. However, what was surprising was that in the majority of the cases, the vegetative height was reduced by on average 2cm, even though the chemicals were not applied until the beginning of flowering. Consequently, the growth of the vegetative internodes does not cease after the start of flowering.

Reproductive Height

The effect of EL500 was to significantly ($p < 0.001$) reduce the reproductive height of the plants by up to 48%. No definite pattern in the effect due to timing of application was evident. JF 10405 reduced ($p < 0.05$), the reproductive height by on average 17%. Again no definite pattern in terms of timing was evident.

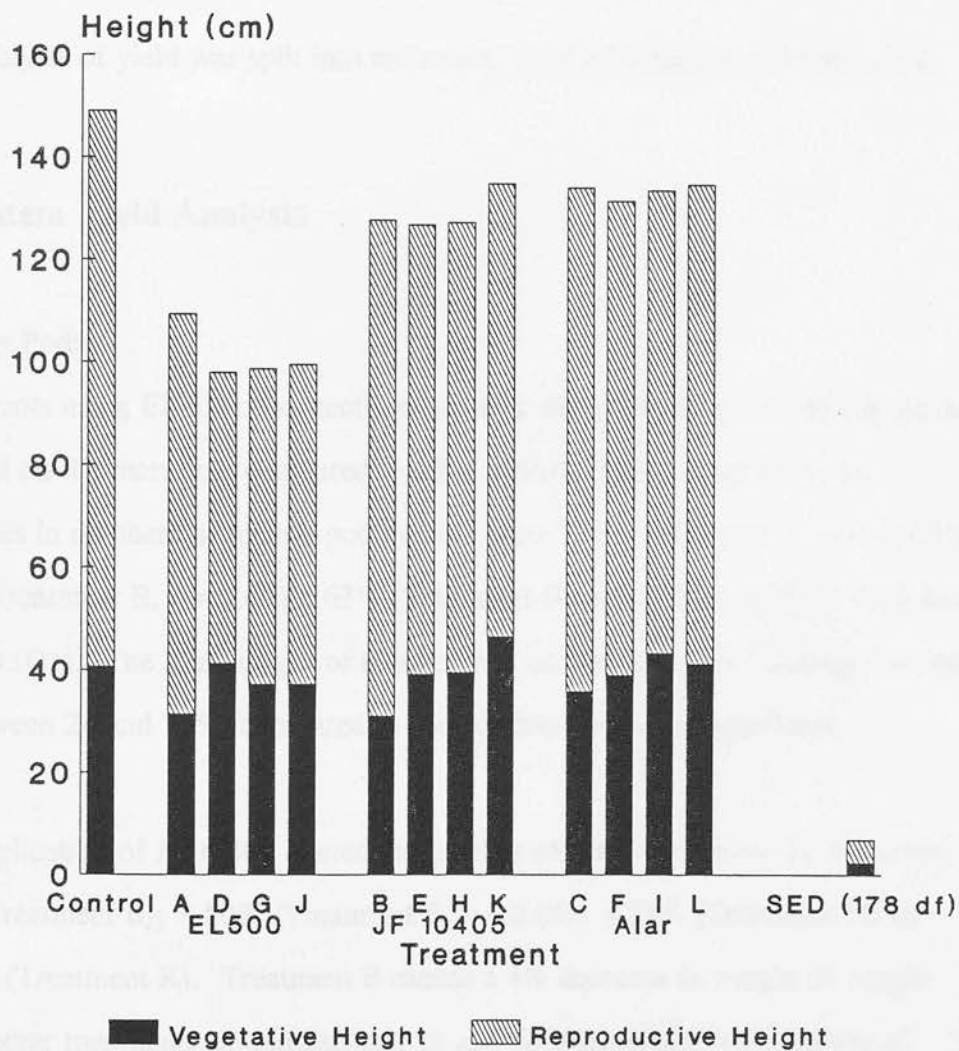


Figure 3.16: Effect of EL500, JF 10405 and Alar, applied at four separate growth stages on vegetative and reproductive height. (Actual figures are shown in Appendix 3.5)

Alar had the least effect upon reproductive plant height at all timings. Reductions (up to 17%), were, however, significant to at least the $p < 0.05$ level in all cases.

Yield Analysis

The analysis of yield was split into mainstem, yield of branches and total yield.

Mainstem Yield Analysis

Mature Pods

Treatments using EL500 consistently resulted in the greatest number of mature pods retained on the mainstem compared to other growth regulator applications.

Increases in numbers of mature pod number were 71% (Treatment A, $p < 0.001$); 74% (Treatment B, $p < 0.001$); 63% (Treatment G, $p < 0.001$) and 89% (Treatment J, $p < 0.001$). The total weight of mature pods on the mainstem, although increased by between 24 and 31% compared to control plants was not significant.

The application of JF 10405 altered the number of mature pods on the mainstem by -9% (Treatment B); +50% (Treatment E, $p < 0.05$); +28% (Treatment H) and +32% (Treatment K). Treatment B caused a 4% decrease in weight of mature pods, other treatments caused between 21 and 54% increases in the weight of mature pods, but these were insignificant.

Alar applied at all growth stages caused more mature pods on the mainstem compared to control plants. Actual increases were 33% (Treatment C); 24% (Treatment F); 37% (Treatment I) and 59% (Treatment L, $p < 0.01$). The weight of mature pods also increased by between 20 and 36%, however, as in all other treatments, these increases were not significant (Table 3.3).

Table 3.3: Effect of EL500, JF 10405 and Alar applied at four separate growth stages on mainstem yield.

Treatment	Number of Mature Pods	Weight of Mature Pods (g)	Number of Harvestable Pods	Weight of Harvestable Pods (g)	Number of Seeds	Weight of Seeds (g)	Weight of Individual Seed (g)	Number of Seeds / Pod
Control	4.6	131.7	4.3	130.6	16.3	36.6	2.2	3.9
A	7.9	172.5	7.3	171.0	28.7	57.0	1.9	4.0
B	4.2	126.6	4.1	126.2	16.1	40.1	2.4	3.8
C	6.1	157.7	5.5	156.4	21.3	42.7	2.0	4.0
D	8.0	163.6	6.5	159.8	25.1	51.7	2.0	3.8
E	6.9	202.5	6.5	201.9	27.9	65.1	2.3	4.2
F	5.7	163.1	5.5	163.0	22.9	53.9	2.4	4.2
G	7.5	164.0	6.3	161.2	24.6	55.7	2.2	4.0
H	5.9	166.5	5.6	159.0	21.1	53.3	2.3	3.8
I	6.3	165.8	5.9	164.3	22.2	53.2	2.4	3.8
J	8.7	158.1	7.3	154.2	26.5	52.8	2.0	3.7
K	6.1	158.9	5.5	157.9	20.7	49.2	2.3	3.8
L	7.3	179.3	6.7	178.0	25.4	56.2	2.2	3.7
SED (178 df)	0.90	24.05	0.81	24.09	3.45	8.15	0.17	0.26
Probability	<0.001	0.258	<0.001	0.266	0.003	0.057	0.010	0.691

All figures represent the mean of 15 plants



Harvestable Pods

Significant increases in the number of harvestable pods were achieved in all cases by the application of EL500. These increases ranged from 47% (Treatment G, $p < 0.01$) to 70% (Treatments A and J, $p < 0.001$). These increases were not as great as the increase in mature pod numbers due to fewer of the retained pods being of a harvestable size. Between 81 and 92% of the mature pods were of a harvestable size in EL500 treatments, compared to 93% in control plants. Total weight of harvestable pods on the mainstem increased by between 18 and 31% however, these were not statistically significant (Table 3.3).

Treatment B decreased the number of harvestable pods (-5%), coupled with a corresponding decrease in weight (-4%). These decreases were not significant. However, it is interesting to note that the proportion of mature pods of a harvestable size was increased from 93% in control plants, to 98%. Other JF 10405 treatments resulted in increases in both harvestable pod numbers (28-51%) and yield (21-55%).

Application of Alar increased the number of harvestable pods on the mainstem by between 28% (Treatment C) and 56% (Treatment L, $p < 0.01$). The number of harvestable pods appeared to increase as timing was delayed. The number of mature pods that were of a harvestable size ranged between 90-96%. This in turn corresponded to increases in the weight of harvestable pods of between 20 and 36%. These increases were not significant.

Seeds

The average number of seeds per pod and average weight of individual seeds on plants treated with EL500 was not altered from control plants. Consequently, the increase in the number of seeds developed on the mainstem was purely due to the increase in the number of harvestable pods. Seed number increased from between 51% (Treatment G, $p < 0.05$) to 76% (Treatment A, $p < 0.001$).

Total weight of seeds per mainstem was increased by the use of EL500 by between 41 % (Treatment D) and 56% (Treatment A), these increases were however, insignificant.

As in the case of EL500, the application of JF 10405 did not affect either the number of seeds per pod or the weight of individual seeds. Treatment B, caused a slight decrease in the weight of harvestable pods, but the weight of seeds on the mainstem was increased by 3.5 %, although the effect was not significant. Other JF 10405 treatments resulted in increased numbers of seeds by between 27% (Treatment K) to 71 % (Treatment E, $p < 0.001$). Increases in weight of seed, although non-significant ranged from 34-78%.

Application of Alar caused the number of seeds developed on the mainstem to be increased by between 31 % (Treatment C) and 56% (Treatment L, $p < 0.05$).

Weight of seeds was increased by between 17% (Treatment C) and 54 % (Treatment L). Again, however, these increases were not significant (Table 3.3).

Branch Yield Analysis

Table 3.4 shows that the application of plant growth regulators had no significant effect on the number of branches on each plant. In addition, application had no significant effect on either the number of harvestable pods, weight of harvestable pods, number of seeds or weight of seeds obtained from the branches.

Overall Yield Analysis

Overall numbers of harvestable pods, weight of harvestable pods, numbers of seeds and weight of seeds was increased by all treatments at all timings, although none of these increases were significant (Table 3.5).

Table 3.4: Effect of EL500, JF 10405 and Alar applied at four separate growth stages on yield of branches.

Treatment	Number of Branches	Number of Harvestable Pods	Weight of Harvestable Pods (g)	Number of Seeds	Weight of Seeds (g)
Control	1.1	3.1	87.5	11.9	25.8
A	1.3	3.3	71.9	11.1	21.9
B	1.5	4.1	123.2	15.5	37.6
C	1.3	3.9	117.7	15.5	31.5
D	1.2	2.9	63.2	9.5	19.5
E	1.6	2.9	82.2	10.6	26.5
F	1.4	4.1	128.8	16.9	40.9
G	1.5	4.2	93.2	13.7	30.0
H	1.4	3.3	106.1	14.0	33.1
I	1.4	3.9	100.1	13.1	32.2
J	1.3	4.9	100.4	18.0	34.9
K	1.1	3.4	80.9	12.2	26.8
L	1.1	3.8	92.6	14.1	30.3
SED (178 df)	0.27	1.11	30.62	4.19	9.64
Probability	0.674	0.877	0.634	0.744	0.676

All figures represent the mean of 15 plants.

Harvest Index

The weight of the pod hull in all treatments except Treatment D, increased compared to the control plants by between 1 and 28%, reflecting the increase in the number of harvestable pods (Table 3.5).

Applications of EL500 to plants decreased stem and leaf fresh weight in all treatments by between 12 and 23% (except Treatment A), corresponding to the

Table 3.5: Effect of EL500, JF 10405 and Alar applied at four separate growth stages on overall yield.

Treatment	Number of Harvestable Pods	Weight of Harvestable Pods (g)	Number of Seeds	Weight of Seeds (g)	Weight of Hull (g)	Weight of Stem + Leaves (g)	Harvest Index
Control	7.3	218.2	28.1	62.3	157.2	218.8	13.8
A	10.6	242.9	39.8	79.0	159.0	220.7	16.9
B	8.2	249.4	31.5	77.7	172.3	241.3	15.9
C	9.4	274.1	36.9	74.2	201.9	260.0	14.2
D	9.4	223.0	34.6	71.2	156.4	169.4	17.9
E	9.5	284.0	38.5	91.6	193.7	274.2	16.3
F	9.7	291.9	39.8	94.7	197.6	230.4	18.2
G	10.5	254.3	38.3	85.7	172.1	186.3	19.6
H	8.9	265.1	35.1	82.6	190.8	234.9	15.4
I	9.8	264.4	35.3	85.4	182.5	235.2	17.1
J	12.2	254.6	44.5	87.7	172.0	193.0	19.2
K	8.9	238.8	32.9	76.0	164.9	244.2	15.6
L	10.5	270.6	39.5	86.5	192.3	240.9	16.2
SED (178 df)	1.59	46.55	6.34	15.26	32.17	36.81	1.36
Probability	0.323	0.942	0.550	0.767	0.911	0.235	<0.001

All figures represent the mean of 15 plants.

decrease in overall plant height. This decrease, coupled to the increases in the overall yield of seed, resulted in an increase in the fresh weight harvest index of 3.1 (Treatment A, $p < 0.05$); 4.1 (Treatment D, $p < 0.01$); 5.8 (Treatment G, $p < 0.001$) and 5.4 (Treatment J, $p < 0.001$).

The fresh weight of stem and leaves was increased (although non-significantly), by the application JF 10405 by between 7% (Treatment H) and 25% (Treatment E), even though the height of these plants was reduced. However, due to increases in yield of seed, the harvest indices were increased by 2.1 (Treatment B); 2.5 (Treatment E); 1.6 (Treatment H) and 1.8 (Treatment K), all non-significant.

As with the application of JF 10405, the application of daminozide caused an increase in the weight of stem and leaves. However, all treatments resulted in an increase in harvest indices of 0.4 (Treatment C); 4.4 (Treatment F, $p < 0.01$); 3.3 (Treatment I, $p < 0.05$) and 2.4 (Treatment L).

Discussion

In general, it was shown that increases in pod set, numbers of mature pods and harvestable pods were more apparent as the timing of application was delayed from flower initiation to growth stage 09. In addition, the ratings of the chemicals in descending order of activity appeared to be EL500, Alar and JF 10405.

The application of EL500 at growth stage 09 had the greatest effect on increased numbers of mature and harvestable pods. This increased number of pods was borne almost exclusively on the mainstem. As the chemical was applied to the apex of the plant, this suggests that either the chemical is not totally mobile within the plant, or

that the chemical is relatively short lived in its activity. Thus, its effects were not available to the later developing flowers of the secondary branches.

Taking the most successful applications as examples (Treatments J, K and L), it can be seen that increases in pod set were evident both within and over all racemes. Thus, the application of the chemical suppressed the plant's inherent dominance towards the lower racemes and proximal flowers. The method by which the chemicals achieve this is unknown, however, it could be suggested that it may be due to a combination of factors. All of the products are described as anti-gibberellins and this was demonstrated by their capacity to decrease stem growth particularly in the reproductive part of the plant by up to 48%. Reduction in stem length was attributed by Attiya *et al.* (1983), as the reason why pod set increased after applying PP333 to the plants. Thus, the plant growth regulator was thought to have increased the ability of pods to compete for assimilates because of a reduction in apical dominance. In addition, however, it can be suggested that pod set is not purely a factor of assimilate availability. The presence or lack of certain hormones can affect the diversion of these assimilates to developing sinks. For example, cytokinins are known to attract assimilates. It may be possible therefore, that the reduction in gibberellins caused by the application of the PGRs resulted in a relative increase in certain intrinsic hormones, e.g. cytokinins and thus pre-disposed the plant to set more pods.

Abou-Elleil and El-Wazeri (1978), stated that the application of daminozide increased flower retention but final yield was reduced due to increased pod drop. In this experiment, a similar phenomena occurred but not to such an extent as to reduce overall yield. In control plants, 51% of the pods that set remained on the plant and developed into harvestable pods. It can be seen from Table 3.2, that as the number of pods set increased, the actual percentage of those pods reaching a harvestable size decreased. In Treatment J for example, only 34% reached this

stage. This may be due either the assimilates needed for pod growth becoming limiting, or the increased competitive effect on the sinks (pods) produced by the application of the PGR gradually wears off. Consequently, it can be seen from the graphs showing mature pod distribution (Figs 3.8, 3.9, 3.10), that the naturally dominant pods gradually resume their status. Much of the reason for this is due to the branching of the vascular tissue within the raceme which favours assimilate supply to proximal pods (Smith, 1982). The majority of mature pods were retained on the lower racemes (Figs. 3.11, 3.12, 3.13), which suggests, that by late pod set, the effects of the anti-gibberellins may have been wearing off. In order to maximise the increased yield potential caused by increased pod set, it may therefore be necessary to either boost assimilate production, or continue to divert assimilates to the pods via longer lasting growth regulators or by using additional applications of plant growth substances. From the distribution of mature pods in Figs 3.8d, 3.9d, and 3.10d, it could be suggested that as proximal dominance was least evident with EL500 treatment, that this chemical had the longest lasting effect. The differences between Alar and JF 10405 are much more difficult to assess, but it could be suggested that Alar is the next most persistent.

Applications of 8 Kg/ha of daminodide at the beginning of flowering increased

Gates *et al.* (1983b), state that yield fluctuations in *Vicia faba* are due primarily to reproductive failure, in particular, to flower drop. Yield increases within this experiment were due in the main to the increase in pod set and pod retention, rather than due to any increases in either average seed size or increased numbers of seeds within each pod. Thus, the results from this experiment support previous findings (Abou-Elleil and El-Wazeri, 1978; Attiya *et al.*, 1983; Smyth, 1986). However, results show, that due to internal competition, as the number of pods increased, the weight of individual seeds generally reduced (albeit insignificantly). This therefore further supports the case for applying either another growth regulator at late pod set or increasing assimilate production in some other way.

Up to 52% yield increases were observed, However, such increases were not significant. The main reason for this was the high degree of variability between plants. In addition, the completely randomized design tended to compound this effect. Thus, it is now known that future experiments should be of a randomized block design. In order to further reduce variability, the experiments could be done on inbred lines, however, the cost involved coupled with the fact that such lines are not commercially available would mean this was not applicable in this thesis.

In conclusion it can be stated that all the chemicals applied resulted in increased yields (albeit insignificant), of broad beans. The most active chemical appeared to be EL500, followed by Alar and JF 10405. The largest increases in pod set resulted from later applications, i.e. at growth stage 09 rather than flower initiation. This tends towards a disagreement with those authors that applied these chemicals during early leaf expansion stages (Attiya *et al.*, 1983; Kellerhals and Keller, 1984). It would be wrong to over state this, however, as a very early application was not made in this experiment.

Applications of 8 Kg/ha of daminozide at the beginning of flowering increased yields by between 10 and 27%, at this application rate, however, the yield increases were considered to be uneconomical (Dekker and Neuval, 1983). In this experiment, much lower levels of daminozide (1.5 Kg/ha) increased yield by up to 52%. It must therefore be concluded that this chemical, EL500 and to a lesser extent JF 10405 warrant further investigation.

Introduction

Chapter 4

Effect of applied growth substances to flowers

Experiments using plant growth regulators (PGRs; Keller and Keller, 1980; Kishimoto and Keller, 1984), as well as those detailed in Chapter 3, have clearly shown that exogenous plant hormones in the initiation of pod set and subsequent development of pods, together with the effects that growth regulators (PGRs) may have upon such hormone concentrations, remains unclear. For example, the yield-enhancing effects of application of PGRs have been variously attributed to a decrease in endogenous concentrations of ABA (El Zewily *et al.*, 1985), or ethylene (Antye *et al.*, 1983). Application of cytokinins in combination with auxin and gibberellin acid to flowers before pollination also resulted in greater pod set (Chapman and Safadi, 1981).

The object of this experiment was to investigate the effects of application to flowers, before and after tripping, of an auxin, a gibberellin or a cytokinin. In order to elucidate the influence of exogenous plant hormones on pod set and development,

Introduction

Experiments using plant growth regulators (Smith, 1982; Keller and Belluci, 1980; Kellerhals and Keller, 1984), as well as those detailed in Chapter 3, have clearly shown that they can significantly increase pod set. However, the role of endogenous plant hormones in the initiation of pod set and subsequent development of pods, together with the effects that growth regulators (PGRs) may have upon such hormone concentrations, remains unclear. For example, the yield-enhancing effects of application of PGRs have been variously attributed to a decrease in endogenous concentrations of ABA (El Zawily *et al.*, 1985), or ethylene (Attiya *et al.*, 1983). Application of cytokinins in combination with auxin and gibberellic acid to flowers before pollination also resulted in greater pod set (Chapman and Sadjadi, 1981).

The object of this experiment was to investigate the effects of application to flowers, before and after tripping, of an auxin, a gibberellin or a cytokinin, in order to elucidate the influence of exogenous plant hormones on pod set and development.

Method

Two seeds of the variety Threefold White were sown into 15cm pots containing Levingtons compost at the Edinburgh School of Agriculture, Bush Estate in February 1988. Within the bee-proof glasshouse, the plants were subjected to a day-time minimum temperature of 21°C and a night-time temperature of 16°C. 400W sodium lamps suspended 1m above the plant and spaced at 1m², provided a 16h photoperiod.

The experiment was arranged as a randomized block, with each treatment replicated 5 times. Every flower on every plant was treated by gently applying solutions of various plant growth substances on to the standard petal and calyx, with a fine artist's paintbrush. Three plant hormones were used: auxin as 4-chloroindole, 3.3×10^{-4} M (50mg/l); cytokinin as 6-benzylaminopurine (BAP), 1×10^{-4} M (23mg/l) and gibberellin as GA₃, 2×10^{-5} M (7mg/l). BAP and GA₃ were first dissolved directly into a few drops of 1M sodium hydroxide and then diluted to strength in distilled water containing 1 ml / litre of the non-ionic wetter "Agral". Chloroindole was directly dissolved into the distilled water and wetter solution.

Plants were treated with the growth substance (Table 4.1) 24h before and 24h after pollination. Pollination was ensured by hand tripping the flowers. Treatments were related to the time of pollination by hand tripping, as the time taken to complete fertilization is difficult to determine and can range from 24 to 72 h after pollination (Stoddard, 1986). Flowers were judged to be at the correct stage for pollination when they had reached flower development stage 9 (Figure 2.2).

Table 4.1: Treatments and times of application of plant growth substance (PGS)

Treatment	Plant Growth Substance	Time of application
Control	Distilled water + Agral	24 h before tripping
A	Gibberellic acid	24 h before tripping
B	Gibberellic acid	24 h after tripping
C	4-chlorindole	24 h before tripping
D	4-chloroindole	24 h after tripping
E	6-benzylaminopurine	24 h before tripping
F	6-benzylaminopurine	24 h after tripping

Plants were scored for pod set, number of mature pods and yield of pods and beans on a per plant basis. In addition, in order to investigate the possible effects of hormone treatment on intra-raceme flower development a detailed analysis of synchrony of flowering was performed. This was calculated by measurement of the time difference, in days, between the proximal flower having fully opened (flower development stage 9) and the distal flower on the same raceme reaching the same stage of development. This figure was divided by the number of flowers on that raceme. The correlation coefficient was calculated between this intra-raceme flowering synchrony figure and the percentage pod set achieved on the same raceme. This procedure was repeated for every raceme formed for each plant in all treatments.

Results

Intra-Raceme Percentage Pod Set

In all control plants the pattern of pod set was the same, most pods set at proximal flower positions on the raceme, especially positions 1 and 2 (Fig. 4.1a, Plate 1). Few pods set at positions more distal than flower 3.

Application of GA₃ before tripping (Treatment A), resulted in plants that showed a similar pattern of pod set to control plants (Fig. 4.1a). However, slightly more pods set (5%), at position 2 than in controls, but this was counterbalanced by a 10% reduction at position 3. Application of GA₃ after pollination (Treatment B), resulted in a significantly ($p < 0.05$), greater pod set at flower position 1 (21%) compared to control plants. This greater pod set was compensated for by a reduction of 2% pod set at flower 2 (Fig. 4.1b).

The application of 4-chloroindole, to flowers 24h before tripping (Treatment C), resulted in an intra-raceme pod set pattern similar to control plants (Fig. 4.2a). Application after pollination (Treatment D), resulted in significantly greater pod set compared to control plants at flower positions 1 (41%, $p < 0.001$); flower 2 (25%, $p < 0.01$) and 4 (12%, $p < 0.05$). The pattern of intra-raceme pod set was maintained, in that most pods set at proximal flower positions and fewer pod set at more distal flower positions.

The application of 6-benzylaminopurine (BAP) to flowers before or after tripping (Treatment E and F), resulted in almost complete pod set (Plate 2) and, in both cases flowers at all positions set significantly ($p < 0.001$) more pods compared to controls (Figs 4.3a and 4.3b) and compared to other treatments.



Plate 1: Distribution of pod set in control plants demonstrating pod set at proximal positions.

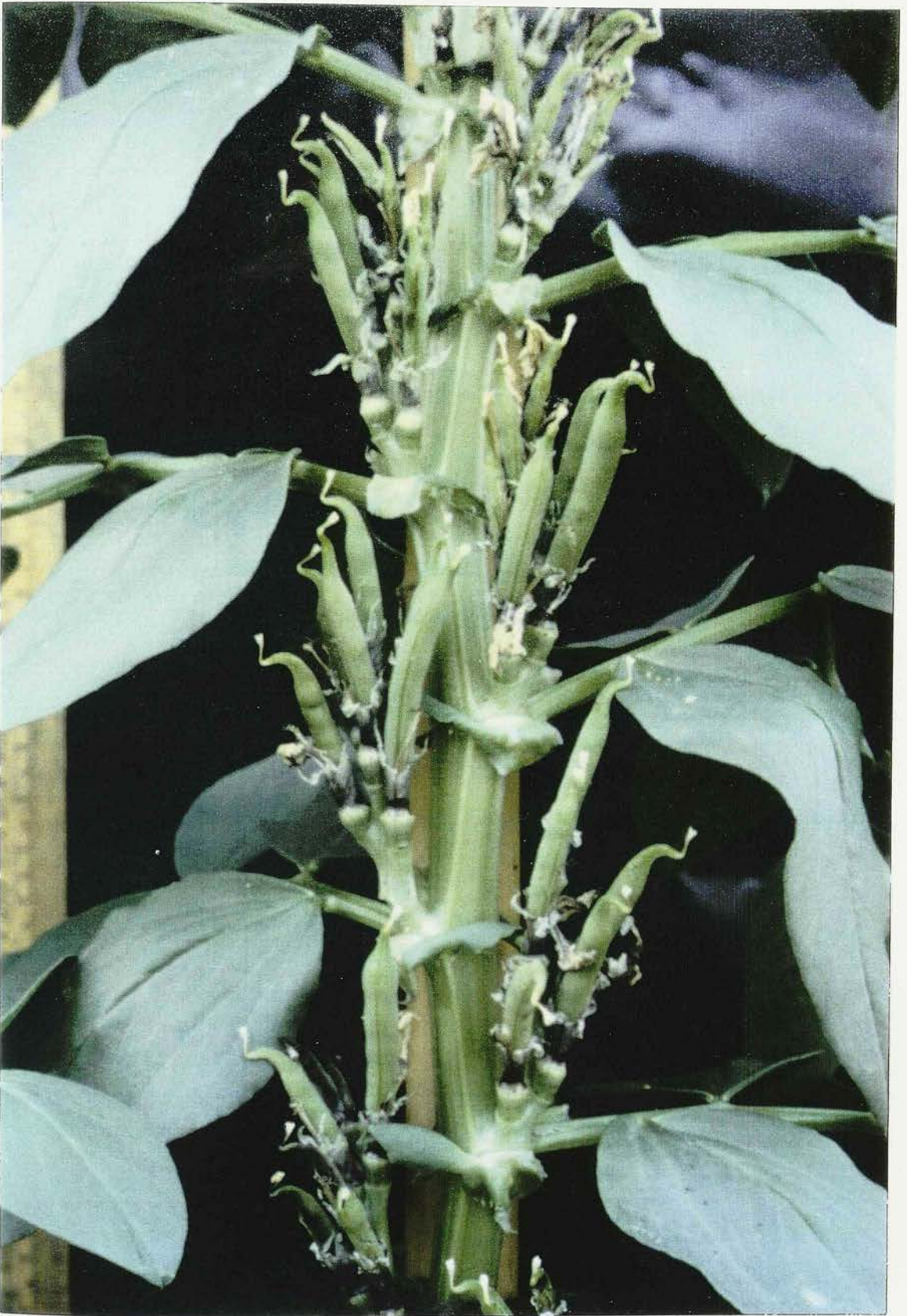


Plate 2: Pod set on plants treated with BAP 24 hours before tripping demonstrating pod set at all flower positions.

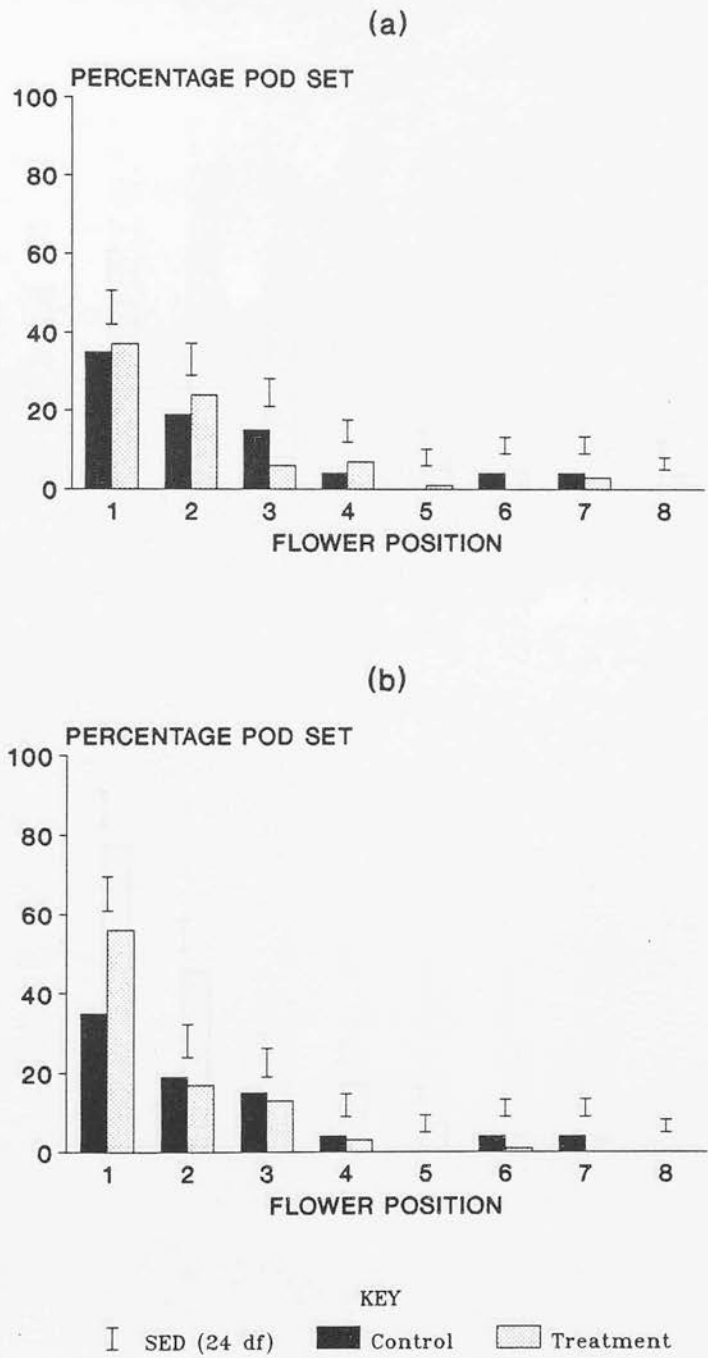


Figure 4.1: Effect of applying GA₃ to flowers a) 24 hours before and b) 24 hours after tripping, on intra-raceme percentage pod set. (Actual figures shown in Appendix 4.1).

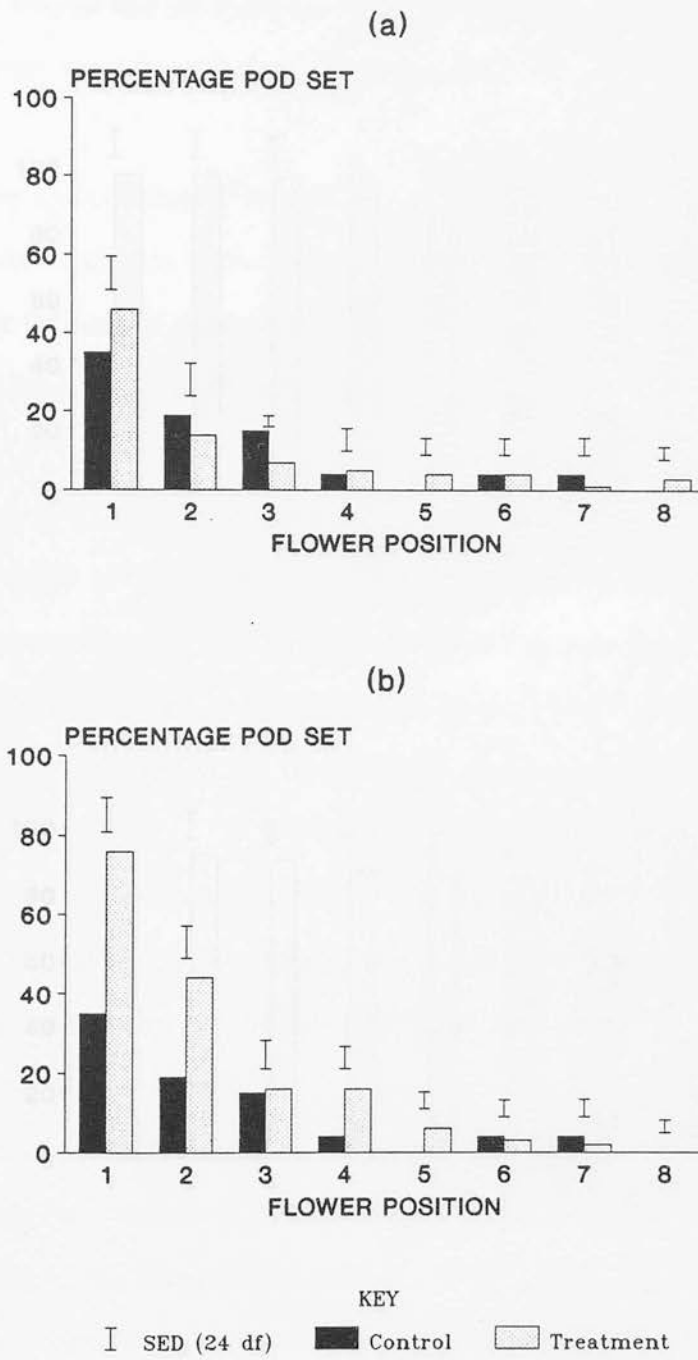


Figure 4.2: Effect of applying 4-chloroindole to flowers a) 24 hours before and b) 24 hours after tripping on intra-raceme percentage pod set. (Actual figures shown in Appendix 4.1).

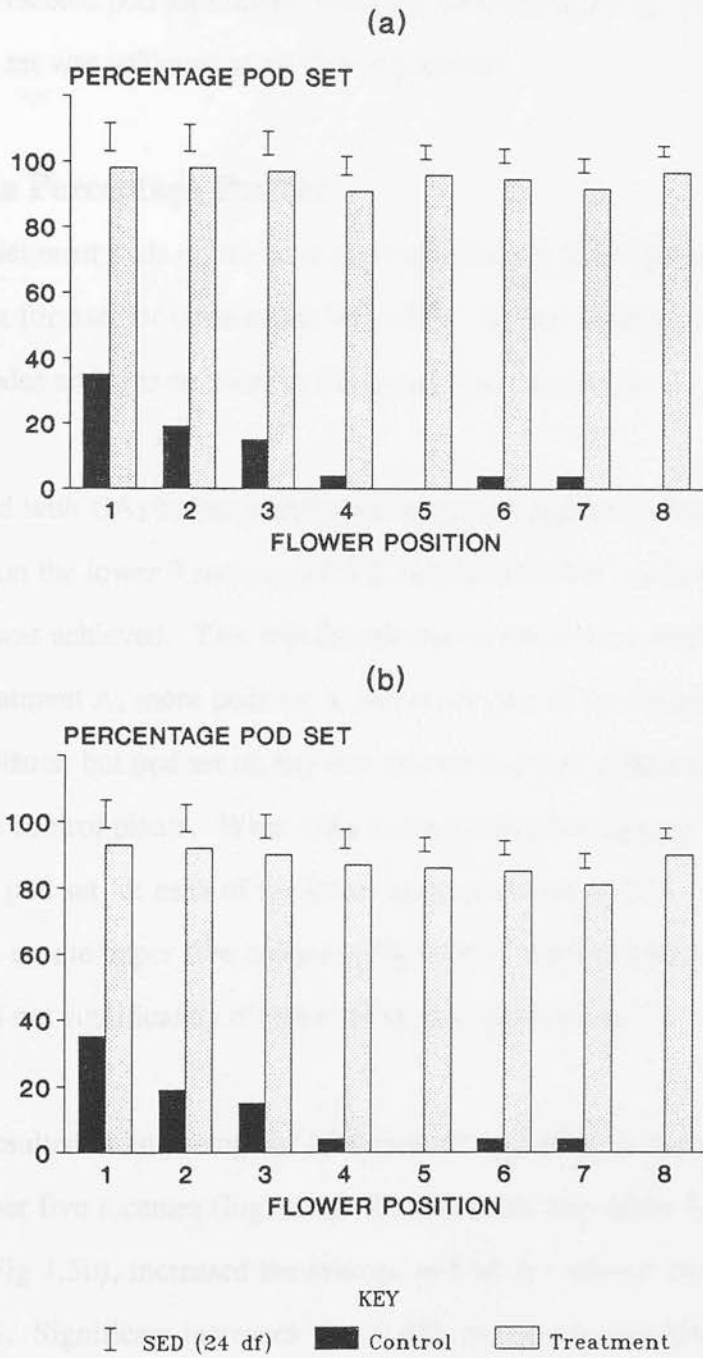


Figure 4.3: Effect of applying BAP to flowers a) 24 hours before and b) 24 hours after tripping on intra-raceme percentage pod set. (Actual figures shown in Appendix 4.1)

Application of BAP before tripping produced 91 - 98% pod set, whereas application after tripping resulted in less and a greater variation of pod set (81 - 93%). The pattern of intra-raceme pod set hitherto observed was not apparent; i.e. a high percentage pod set was achieved at all flower positions.

Inter-Raceme Percentage Pod Set

Control plants set most pods on the first nine reproductive nodes formed (Fig 4.4a). Average pod set for each of these nodes was 13%. Pod set declined towards apical reproductive nodes and was on average 3% on each of racemes 9-14.

Flowers brushed with GA₃ before tripping (Treatment A), produced an average pod set per raceme on the lower 9 racemes of 9%, whereas for the remaining five racemes, 18% was achieved. This was the reverse of the control plants (Fig. 4.4a). Thus, after Treatment A, more pods set in the upper part of the reproductive portion of the plants, but pod set on any one raceme was not significantly different from that of the control plants. When GA₃ was applied after tripping (Treatment B), the average pod set for each of the lower nine racemes was 16%, with an average of 10% on the upper five racemes (Fig 4.4b). Again, however, pod set on any raceme was not significantly different from that achieved by the control plants.

Treatment C, resulted in an average of 13% pod set on the lower nine racemes and 10% on the upper five racemes (Fig 4.5a). Chloroindole applied to flowers 24 h after tripping (Fig 4.5b), increased the average pod set for each of the first nine racemes to 29%. Significant increases ($p < 0.05$), in pod set were achieved at the lower four racemes. Average pod set on the upper five racemes was 30%. However, pod set was only significant ($p < 0.05$), compared to control plants on racemes 11, 12 and 13.

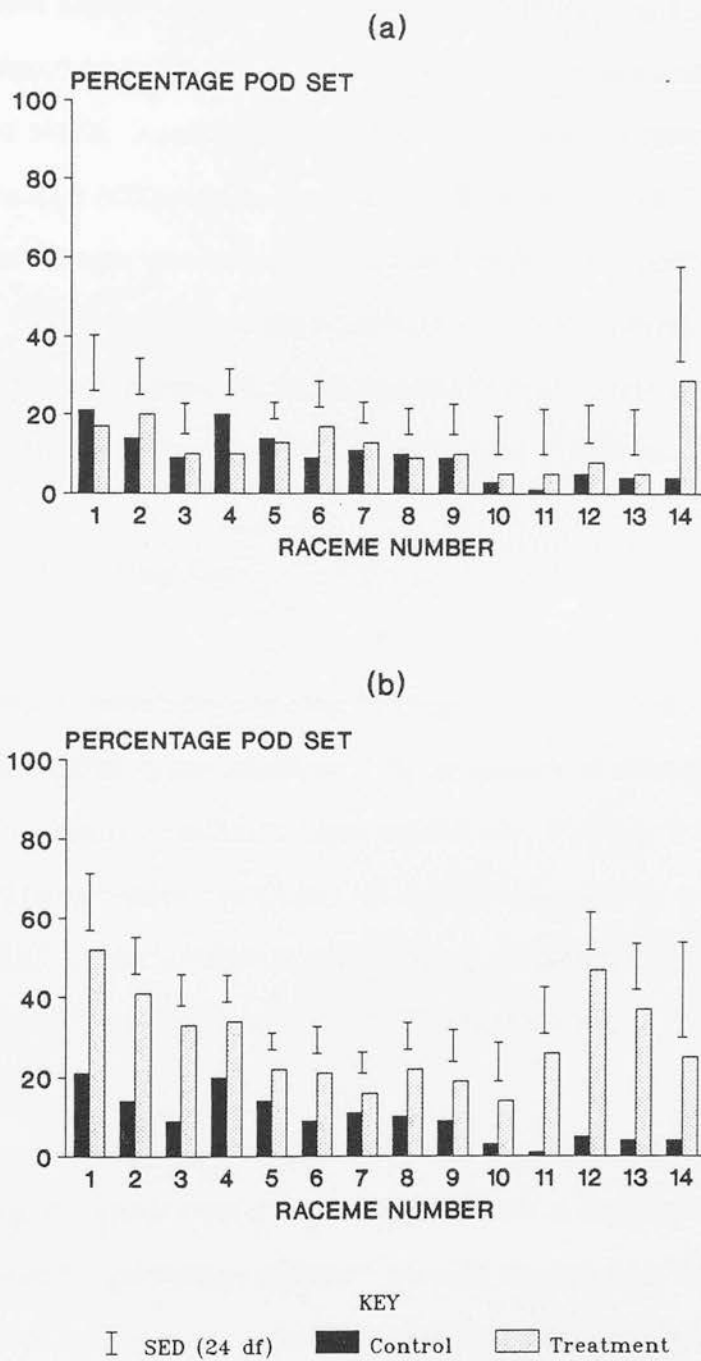
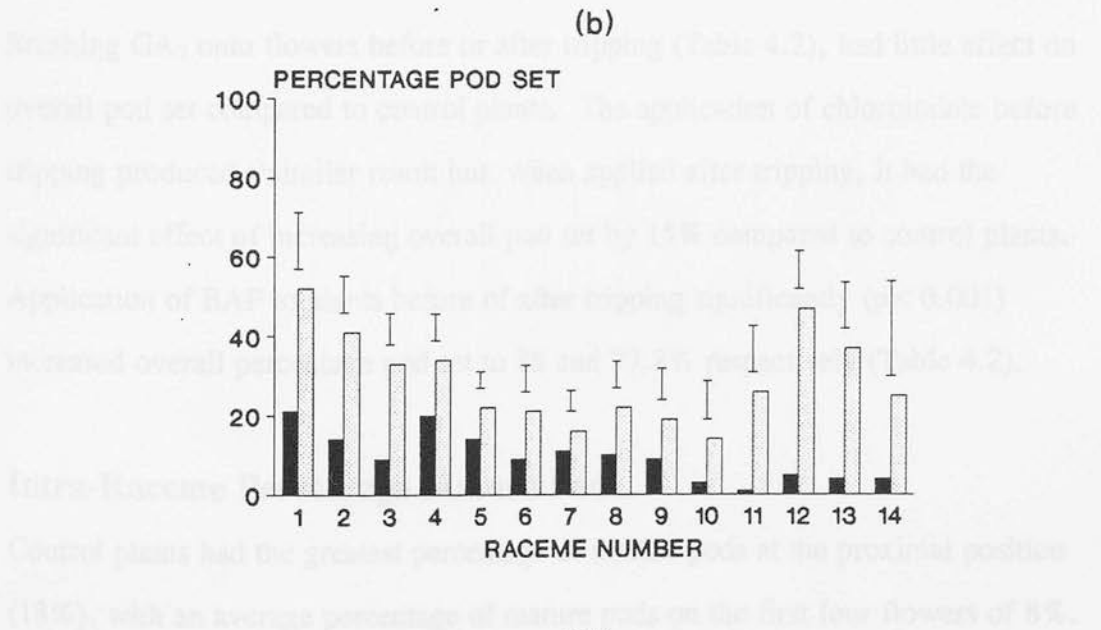
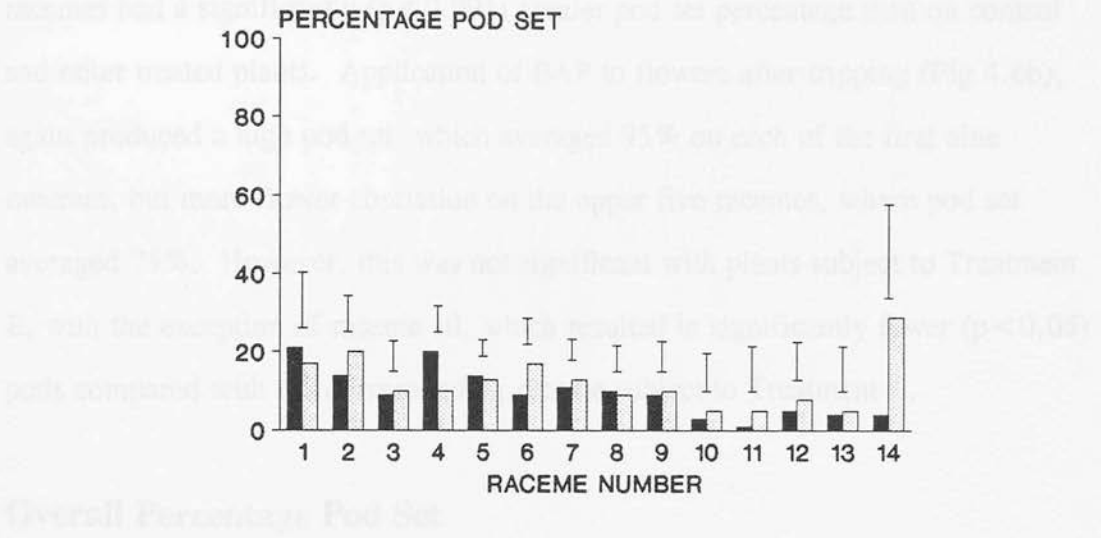


Figure 4.4: Effect of applying GA₃ to flowers a) 24 hours before and b) 24 hours after tripping on percentage inter-raceme pod set. (Actual figures shown in Appendix 4.2)

The application of RAI to flowers before tripping (Treatment B), influenced in almost complete pod set on all racemes, with little difference between the lower part of the reproductive region (91.5%) and the upper portion (98%, Fig. 4.5a). All racemes had a similar pod set, with the lower part of the raceme having a slightly higher pod set than the upper part.



Control plants had the greatest percentage of mature pods at the proximal position (11%), with an average percentage of mature pods on the first four flowers of 65%. Average percentage of mature pods on the first four flowers was 3.25% (Fig. 4.7a). Application of humic acid to the flowers had no effect on the percentage of mature pods on any flower position, even in flower 3. At this flower

Figure 4.5: Effect of applying chloroindole a) 24 hours before and b) 24 hours after tripping on inter-raceme percentage pod set. (Actual figures shown in Appendix 4.2)

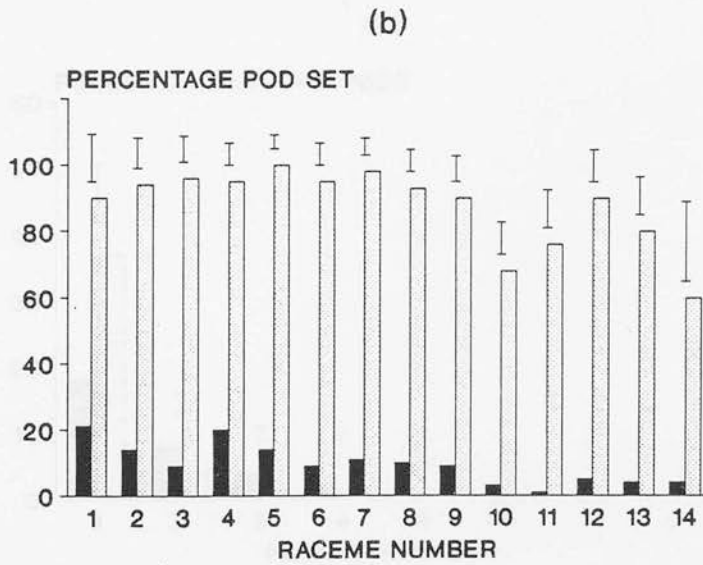
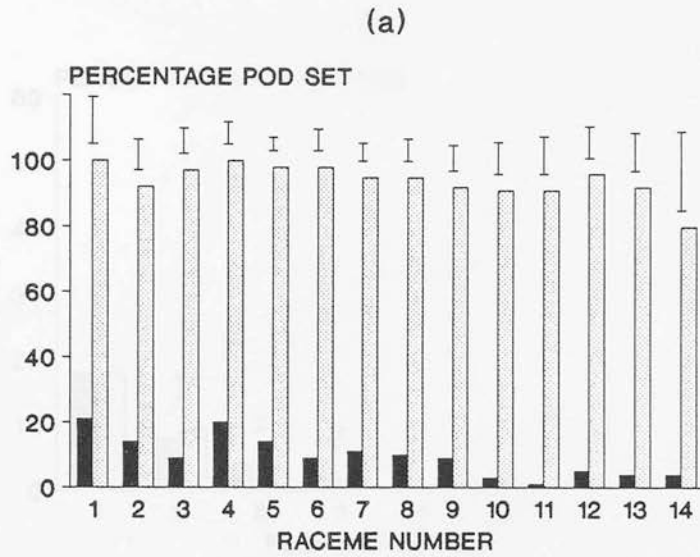
The application of BAP to flowers before tripping (Treatment E), culminated in almost complete pod set on all racemes, with little difference between the lower part of the reproductive region (96%) and the upper portion (90%, Fig 4.6a). All racemes had a significantly ($p < 0.001$) greater pod set percentage than on control and other treated plants. Application of BAP to flowers after tripping (Fig 4.6b), again produced a high pod set, which averaged 95% on each of the first nine racemes, but more flower abscission on the upper five racemes, where pod set averaged 75%. However, this was not significant with plants subject to Treatment E, with the exception of raceme 10, which resulted in significantly fewer ($p < 0.05$) pods compared with the corresponding raceme subject to Treatment F.

Overall Percentage Pod Set

Brushing GA_3 onto flowers before or after tripping (Table 4.2), had little effect on overall pod set compared to control plants. The application of chloroindole before tripping produced a similar result but, when applied after tripping, it had the significant effect of increasing overall pod set by 15% compared to control plants. Application of BAP to plants before or after tripping significantly ($p < 0.001$) increased overall percentage pod set to 85 and 77.8% respectively (Table 4.2).

Intra-Raceme Percentage Mature Pods

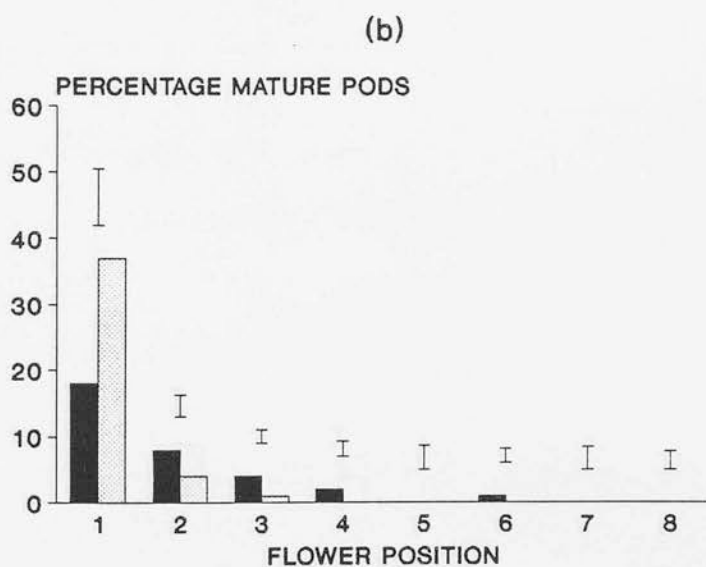
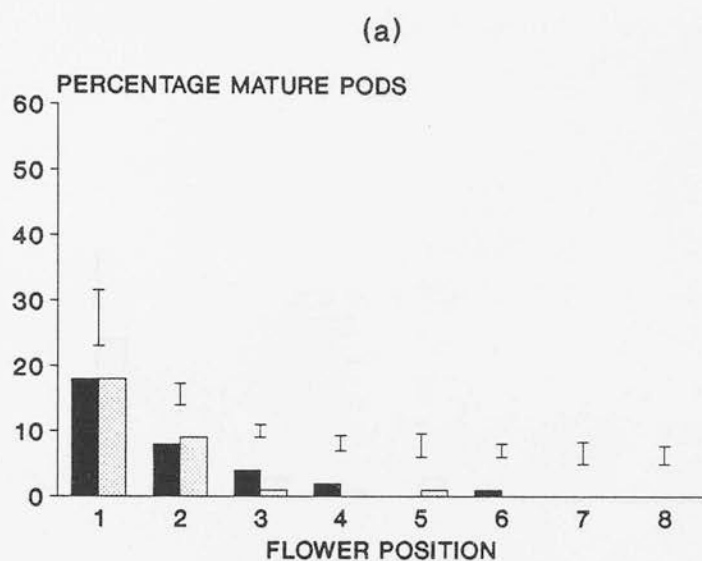
Control plants had the greatest percentage of mature pods at the proximal position (18%), with an average percentage of mature pods on the first four flowers of 8%. Average percentage mature pods on the distal four flower positions was 0.25% (Fig. 4.7a). Application of hormones to the flowers had no effect on the percentage of mature pods retained at any flower position, apart from flower 5. At this flower position, the application of BAP pre-tripping increased percentage mature pods to 9% ($p < 0.05$, Fig 4.9a) and post tripping to 14% ($p < 0.001$, Fig 4.9b).



KEY

┌ SED (24 df) ■ Control □ Treatment

Figure 4.6: Effect of applying BAP to flowers a) 24 hours before and b) 24 hours after tripping on inter-raceme percentage pod set. (Actual figures shown in Appendix 4.2)



KEY

┌ SED (24 df) ■ Control ▨ Treatment

Figure 4.7: Effect of applying GA₃ to flowers a) 24 hours before and b) 24 hours after tripping on intra-raceme percentage mature pods. (Actual figures shown in Appendix 4.3)

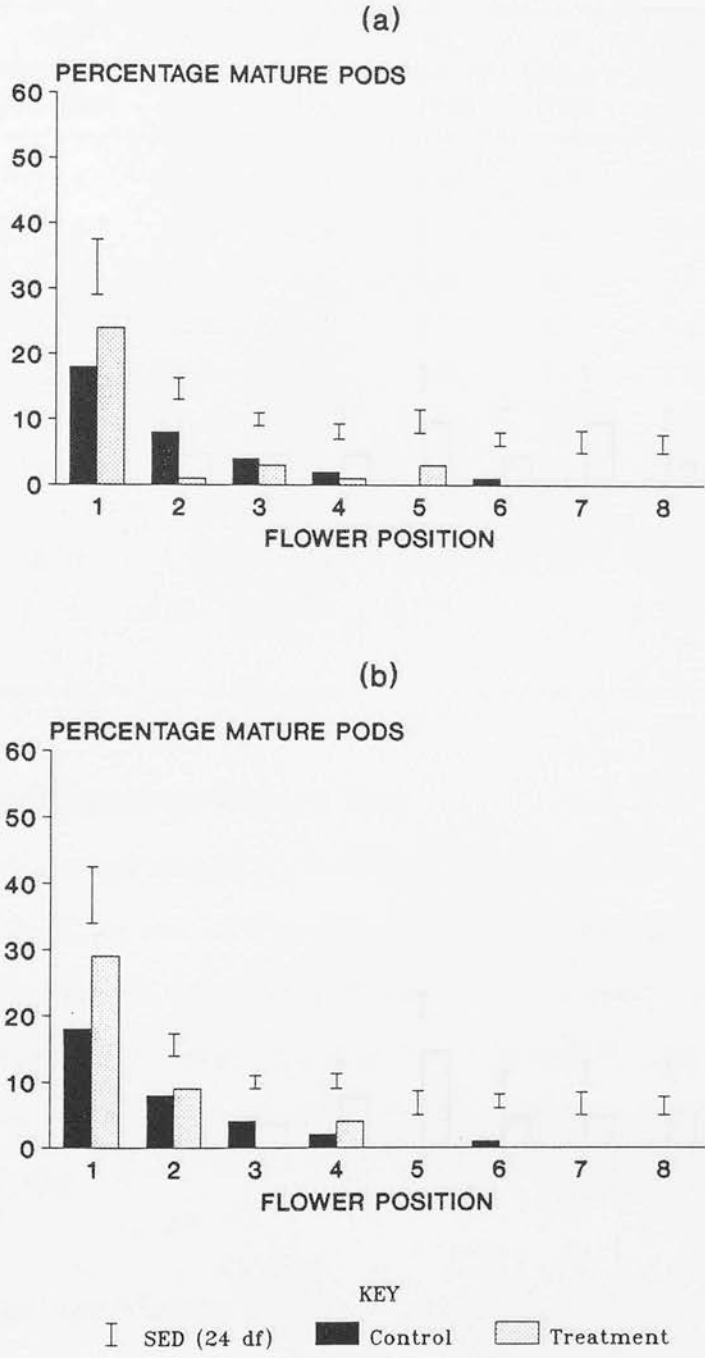


Figure 4.8: Effect of applying chloroindole a) 24 hours before and b) 24 hours after tripping on intra-raceme percentage mature pods. (Actual figures shown in Appendix 4.3)

Table 4.2: Effect of applied plant growth substances on overall percent and mature pods

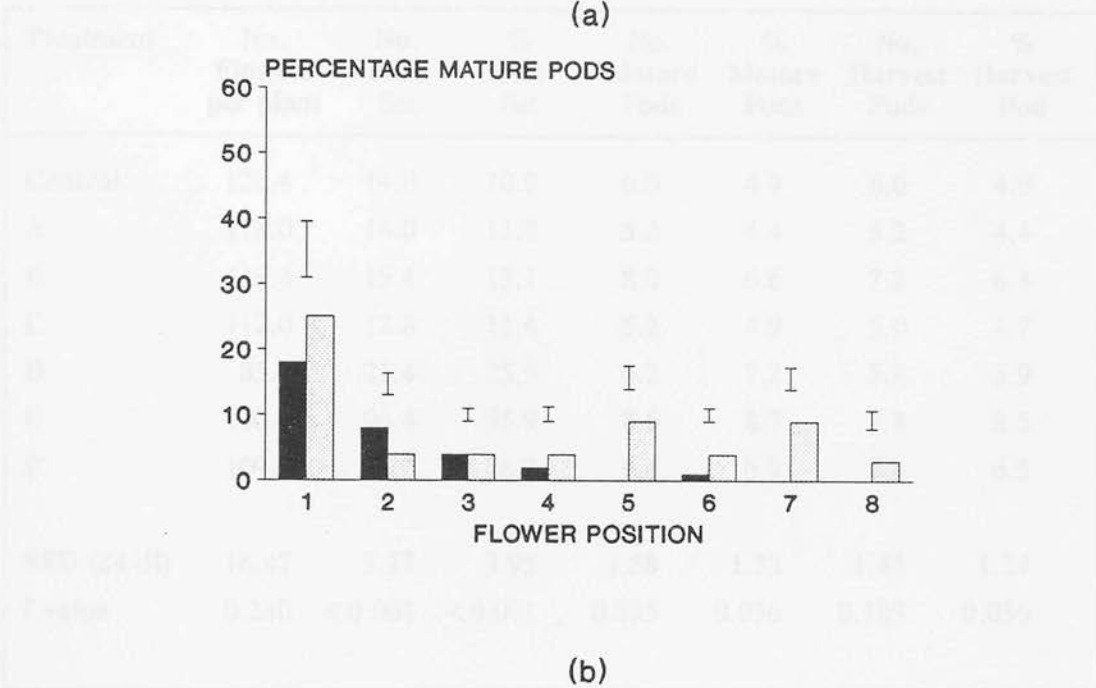


Figure 4.9: Effect of applying BAP a) 24 hours before and b) 24 hours after tripping on intra-raceme percentage mature pods. (Actual figures shown in Appendix 4.3)

Table 4.2: Effect of applied plant growth substances on overall pod set and mature pods

Treatment	No. Flowers per plant	No. Pods Set	% Pod Set	No. Mature Pods	% Mature Pods	No. Harvest Pods	% Harvest Pod
Control	128.4	14.0	10.9	6.0	4.9	6.0	4.9
A	118.0	14.0	11.8	5.2	4.4	5.2	4.4
B	119.6	15.4	13.1	8.0	6.6	7.8	6.4
C	112.0	12.8	11.4	5.2	4.9	5.0	4.7
D	85.6	21.4	25.9	6.2	7.2	5.8	6.9
E	100.6	96.4	95.9	8.6	8.7	7.8	8.5
F	109.8	97.0	88.7	8.4	6.9	7.4	6.5
SED (24 df)	16.47	5.77	3.95	1.58	1.32	1.45	1.24
f value	0.240	<0.001	<0.001	0.235	0.036	0.189	0.036

ALL FIGURES REPRESENT THE MEAN OF 5 PLANTS

Inter-Raceme Percentage Mature Pods

Control plants on average retained 5.1% mature pods on each of the lower nine racemes, while the figure was 1.8% on the upper five racemes (Fig 4.10a). The application of plant growth substances to the flowers had no significant effect on the inter-raceme percentage of mature pods apart from at raceme 9. The application of GA₃ post-tripping increased the percentage of mature pods at raceme 9 to 16% ($p < 0.01$, Fig 4.10b).

Overall Percentage Mature Pods

Control plants retained an average of 6 mature pods per plant (Table 4.2), which represented a percentage mature pod figure of 4.9%. Application of hormones to the flowers had no effect on the number of mature pods per plant. However, the application of BAP pre-tripping resulted in a significant increase ($p < 0.05$) in the percentage of flowers that retained mature pods.

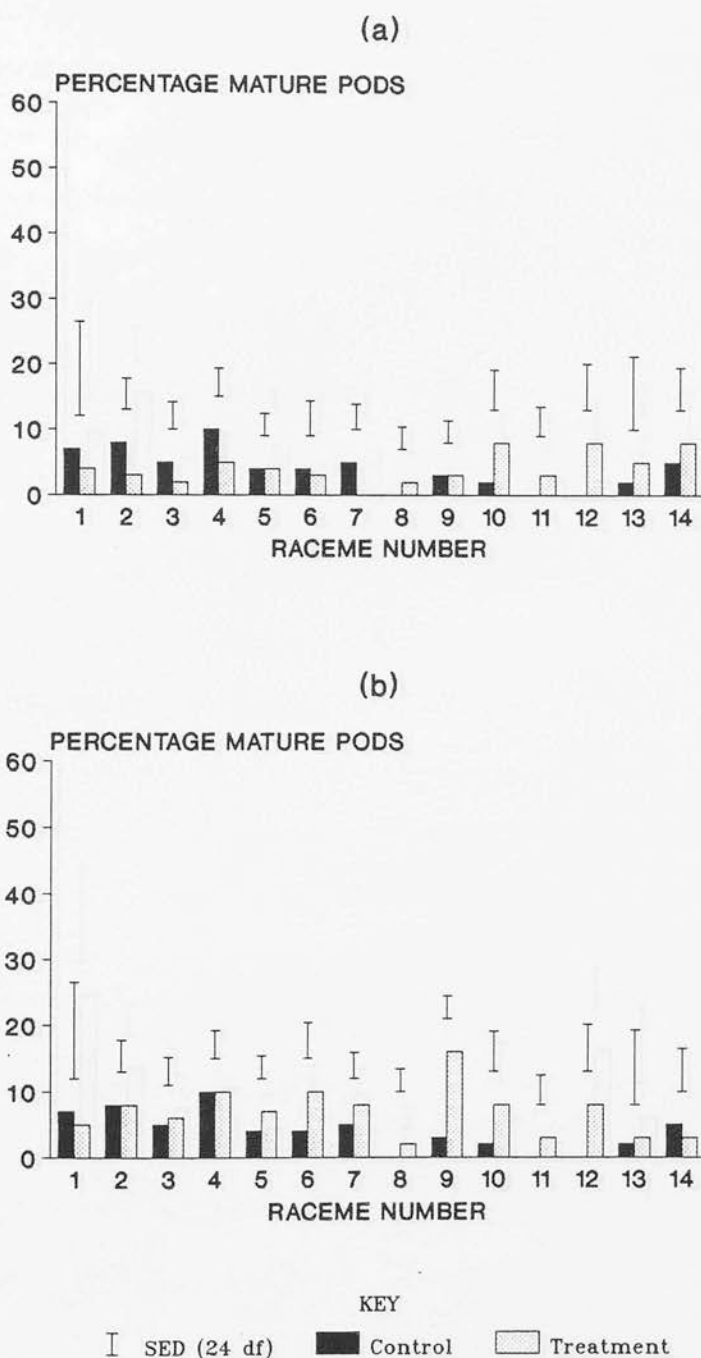
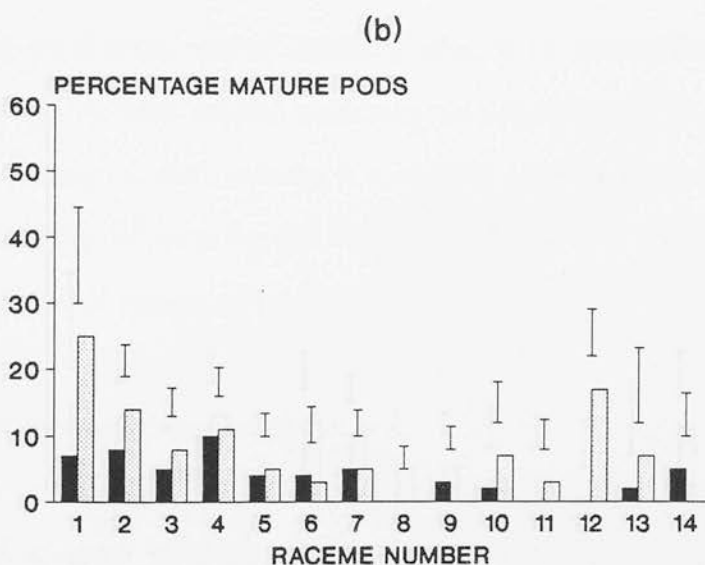
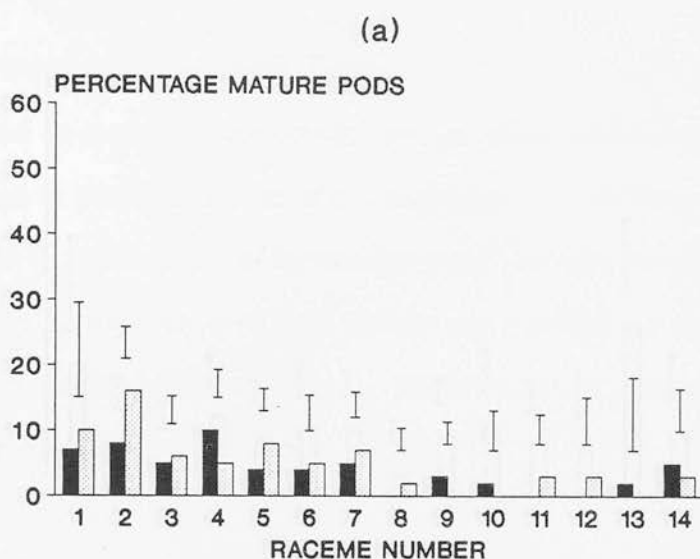


Figure 4.10: Effect of applying GA3 a) 24 hours before and b) 24 hours after tripping on inter-raceme percentage mature pods. (Actual figures shown in Appendix 4.4)



KEY

┌ SED (24 df) ■ Control □ Treatment

Figure 4.11: Effect of applying chloroindole a) 24 hours before and b) 24 hours after tripping on inter-raceme percentage mature pods. (Actual figures shown in Appendix 4.4)

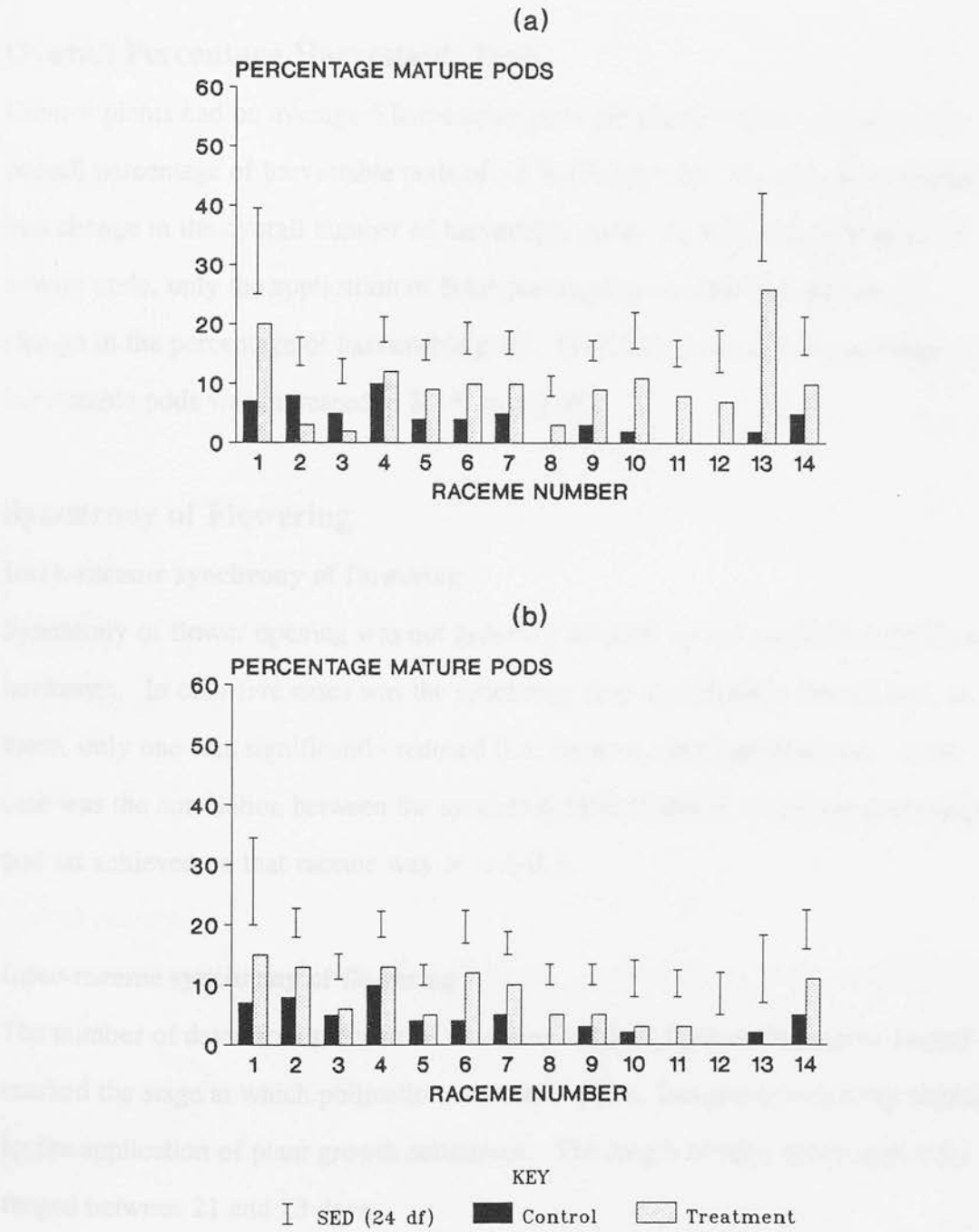


Figure 4.12: Effect of applying BAP a) 24 hours before and b) 24 hours after tripping on inter-raceme percentage mature pods. (Actual figures shown in Appendix 4.4)

No other treatment resulted in significant changes in the percentage of mature pods retained.

Overall Percentage Harvestable Pods

Control plants had on average 6 harvestable pods per plant, which represented an overall percentage of harvestable pods of 4.9% (Table 4.2). No treatment resulted in a change in the overall number of harvestable pods. As with the percentage of mature pods, only the application of BAP pre-tripping resulted in a significant change in the percentage of harvestable pods. With Treatment E the percentage of harvestable pods was increased to 8.5% ($p < 0.05$).

Synchrony of Flowering

Intra-raceme synchrony of flowering

Synchrony of flower opening was not generally affected by the application of plant hormones. In only five cases was the synchrony ratio significantly altered and, of these, only one was significantly reduced (i.e. became more synchronous). In no case was the correlation between the synchrony ratio (Table 4.3) and the percentage pod set achieved on that raceme was $> \pm 0.5$.

Inter-raceme synchrony of flowering

The number of days from the start of flowering until all flowers on raceme 14 had reached the stage at which pollination could take place, was not significantly altered by the application of plant growth substances. The length of time in all cases only ranged between 21 and 23 days.

Table 4.3: Effects of application of plant growth substances to each flower on the intra-raceme synchrony of flowering

Raceme No.	Treatment							SED (24 df)
	Control	A	B	C	D	E	F	
1 (base)	0.82	0.83	0.80	0.75	0.85	0.83	0.86	0.154
2	0.58	0.57	0.62	0.51	0.68	0.54	0.61	0.125
3	0.44	0.42	0.51	0.42	0.43	0.39	0.38	0.066
4	0.38	0.40	0.50	0.50	0.38	0.46	0.33	0.062
5	0.38	0.42	0.48	0.43	0.38	0.60	0.38	0.063
6	0.44	0.50	0.43	0.44	0.52	0.52	0.46	0.087
7	0.44	0.43	0.60	0.61	0.61	0.52	0.49	0.086
8	0.53	0.48	0.56	0.48	0.58	0.56	0.42	0.087
9	0.56	0.41	0.60	0.51	0.51	0.61	0.38	0.088
10	0.60	0.65	0.54	0.52	0.80	0.63	0.50	0.158
11	0.48	0.55	0.61	0.61	0.71	0.61	0.66	0.133
12	0.60	0.52	0.60	0.63	0.82	0.74	0.55	0.135
13	0.73	0.56	0.61	0.61	0.84	0.62	0.61	0.152
14 (apex)	0.48	0.78	0.50	0.49	0.64	0.78	0.47	0.140
Correlation coefficient between synchrony and mean pod set on all racemes	-0.05	0.13	0.26	-0.14	0.40	0.32	-0.20	

Yield Analysis

The yield of treated plants was confined to the mainstem (Table 4.4). Control plants on average retained 6 harvestable pods weighing 124.5g. Within these pods there were 16 seeds which weighed a total of 45.7g. The application of plant growth substances did not significantly affect number of harvestable pods, weight of harvestable pods, number of seeds or the weight of seeds per mainstem.

Table 4.4: Effect of applied plant growth substances to the flowers on mainstem yield

Treatment	Number Mature Pods	Number Harvest Pods	Weight Harvest Pods (g)	Number Seeds	Weight Seeds (g)
Control	6.0	6.0	124.5	16.0	45.7
A	5.2	5.2	112.2	13.6	38.6
B	8.0	7.8	150.5	27.0	66.4
C	5.2	5.0	112.7	19.6	47.0
D	6.2	5.8	97.8	15.6	39.1
E	8.6	7.8	146.4	21.8	55.3
F	8.4	7.4	141.1	23.6	52.1
SED (24 df)	1.58	1.45	27.24	5.06	12.20
f value	0.235	0.189	0.402	0.138	0.304

Reproductive Plant Height

The application of plant growth substances to each flower had no significant effect on the reproductive height of the plants (Fig. 4.13). However, there appeared to be a trend in the heights, whereby pre-pollination applications were slightly taller than post-pollination treatments. Gibberellin and auxin treatments appeared in general, to be taller than either control or BAP treated plants.

Figure 4.13: Effect of applied plant growth substances to each flower on the reproductive height. (Actual figures are shown in Appendix 4.3)

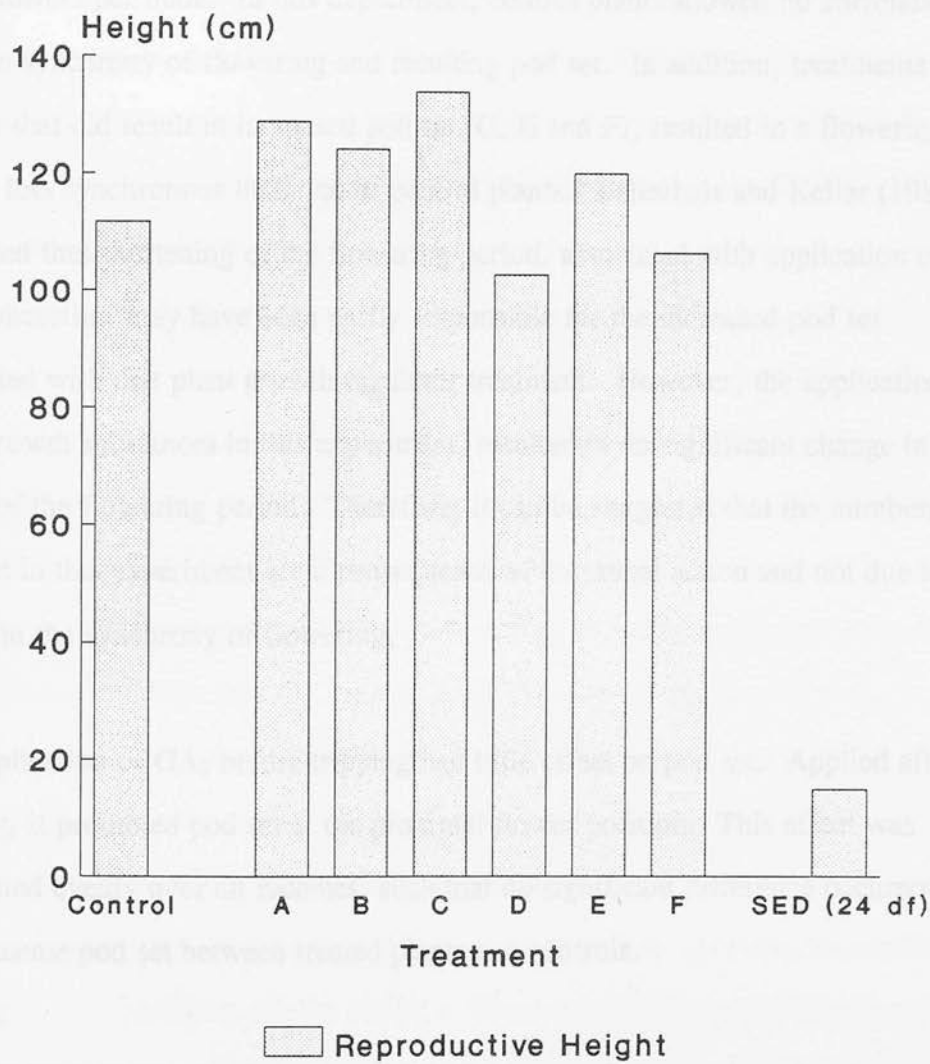


Figure 4.13: Effect of applied plant growth substances to each flower on the reproductive height. (Actual figures are shown in Appendix 4.5)

Discussion

Gates *et al.* (1981), suggested that inbred lines that develop few flowers per node and all opening within 1 - 2 days are less likely to shed their flowers than those with many flowers per node. In this experiment, control plants showed no correlation between synchrony of flowering and resulting pod set. In addition, treatments to flowers that did result in increased pod set (C, E and F), resulted in a flowering pattern less synchronous than that of control plants. Kellerhals and Keller (1984), suggested that shortening of the flowering period, associated with application of an anti-gibberellin, may have been partly responsible for the increased pod set associated with that plant growth regulator treatment. However, the application of plant growth substances in this experiment, resulted in no significant change in the length of the flowering period. Therefore, it can be suggested that the numbers of pods set in this experiment are a consequence of hormonal action and not due to any effects in the synchrony of flowering.

The application of GA₃ before tripping had little effect on pod set. Applied after tripping, it promoted pod set at the proximal flower position. This effect was distributed evenly over all racemes, such that no significant difference occurred in inter-raceme pod set between treated plants and controls.

It would be wrong to attribute the results of this experiment to the role of endogenous cytokinins. It does appear, however, that exogenous application of cytokinins can promote almost complete pod set and it can be postulated that similar intrinsic chemicals may be extremely important in the process of pod set.

It is well established that cytokinins can promote cell division in the vascular tissue, but that cytokinins are inactive in the absence of auxin (Moore 1979). In this experiment, auxin was presumably present intrinsically, although perhaps in

limiting concentrations, so the application of BAP to flowers resulted in active cell division of the embryo and hence the attraction of assimilates to the new developing pods from other plant parts. This view is supported by results obtained by Chapman and Sadjadi (1981), where benzyl-adenine was applied exogenously to distal flowers only. In their case, treatment of distal flowers caused more pods to set at the expense of untreated ones. Thus, exogenous cytokinins appear to be able to promote the formation of, or perhaps aid the preparation of, potentially active reproductive sinks which are rapidly established after the stimulus of pollination and can then effectively divert resources from competing sinks, primarily the vegetative apex and stem and from sources such as the leaves.

In peas (*Pisum sativum*), the cytokinins obtained in the pod development phase before embryo growth, resemble those exported from the root system (Burrows and Carr, 1970). In *Vicia faba*, lower concentrations of cytokinin may be transported from the roots to other plant parts during flowering, possibly because of reduced root growth during this period. Such reduced root growth may be, in part, responsible for flower abscission. Lesina (1966) showed that the application of GA_3 reduced the weight of roots in *V. faba*. Consequently, the opposite effect may occur after the application of anti-gibberellin-type plant growth retardants, i.e. an increase in root weight may be observed, increasing in turn cytokinin production so resulting in increased pod set. In addition, Henson and Wheeler (1976), discovered that root nodules in *V. faba* have high intrinsic levels of cytokinin. The anti-gibberellin compound chlormequat, when applied to field beans (Hassan and El-Moursi, 1982), increased nodulation. Enhanced pod set associated with the application of anti-gibberellins in Chapter 3, may therefore be partly attributed to an increase in cytokinin concentrations due to greater nodulation.

The change in pattern of flower drop and pod set exhibited by plants supplied with auxin and to an extent gibberellin, from before to after tripping (Figs 4.1, 4.2, 4.4

and 4.5), suggests that an "hormonal switch" occurs in terms of auxin and gibberellin requirement at, or directly after, tripping. Before tripping, reproductive organs are insensitive to auxin and gibberellin application; after tripping, greater pod set results. With regard to auxin, Engvild (1985), showed that chloroindole concentrations in developing field bean seeds decreases initially after fertilization. However, in peas, a species that does not suffer from appreciable flower drop (Richards and Smith, 1987), no initial decline in chloroindole was observed; instead, concentrations increased by $> 10,000$ times in the first 20 days after fertilization. Gates *et al.* (1983b) stated that in *V. faba* a lag period prior to active pod growth coincided with vascular differentiation at the pedicel-peduncle junction. Auxin is known to play a major role in vascular differentiation (Bandurski and Nonhebel, 1984). Thus the observed increase in pod set displayed by application of chloroindole after tripping can be attributed to the redirection of assimilates primarily to the pedicel-peduncle junction to facilitate vascular differentiation in that region. This role appears to be similar to that of cytokinin, except that cell expansion is probably promoted by auxin application (Moore, 1979). This was particularly true of flowers at proximal positions in the raceme, where it may be supposed that cytokinin supply was not limiting.

It appears, however, that both auxin and cytokinin and to a certain extent gibberellin, in combination, have a crucial role in the process of pod set in *V. faba*. It is possible that the stimulus to pod set referred to by Gates *et al.* (1981), may be auxin based. The auxin gradient theory of abscission (Addicott *et al.* 1955), states that abscission is induced when the proximal:distal ratio across a potential abscission zone, in this case at the base of the flower, is increased. Diethelm *et al.* (1986), stated that the concentration of hormones within a flower is slightly reduced between bud initiation and full flower development. Thus, if a flower distal within a raceme is considered, it can be suggested that the lowering of internal hormone concentrations, compared with increases in an auxin-based stimulus proximal to it,

originating from pollinated flowers and the apex, may induce the cellular processes leading to abscission, whether the flower is fertilized or not. This theory is supported by Tamas *et al.* (1986), who discovered that the removal of older pods in *Phaseolus vulgaris*, decreased the abscission rate of younger pods, however, this effect was reversed when excised seeds were replaced with IAA or NAA.

Cytokinins, however, may be more important in the establishment of reproductive sinks, whereas auxins and gibberellins may be involved after fertilization in their maintenance and further development.

Increased pod drop associated with all treated plants resulted in similar levels of harvestable pods on both control and treated plants. This suggests that either a) auxins and gibberellins become limiting later in the growth of the plant; or b) the amount of assimilates generated by the plant become limiting. It can be seen from Figs. 4.7 - 4.12, that the distribution of these harvestable pods reverts to the **NORMAL** distribution of mature pods i.e. at proximal flower sites on lower racemes. It would appear logical therefore, to either increase the amount of gibberellin or auxin supply, or to increase assimilate production of the plant in order to harness the increased yield potential stimulated by the application of cytokinins.

Introduction

Chapter 5

Effect of plant growth retardants on dry matter distribution

The aim of this experiment was to apply the plant growth regulators (at the same growth stage as already determined as being the most advantageous in Chapter 3), and to monitor the growth of the treated plants by measuring the composition, with a view to providing a more detailed knowledge of the effects of each substance on plant growth and provide some evidence to support the above hypothesis.

Method

Two seeds of the variety *Thousand White* were sown into 15cm pots containing Levington peat compost and perlite during January 1979 in a box-glasshouse at the Bush Estate, Edinburgh School of Agriculture. Seedlings were thinned and staked to prevent lodging. The plants were subjected to minimum day-time and night-time temperatures of 21 and 14°C respectively. Supplementary

Introduction

It was suggested in Chapter 4, that cytokinins are involved in the establishment of reproductive sinks, hence, leading to increased levels of pod set (Rylott and Smith, 1990). Field trials conducted by ^{the} authors outlined in Chapter 3, have shown that plant growth regulators e.g. Alar, JF 10405 and EL500 can increase pod set in *Vicia faba*. The application of anti-gibberellin plant growth regulators also reduces plant height. These chemicals may in turn cause an increase in the proportion of roots to shoots and as a result increase cytokinin production (Rylott and Smith, 1990). Root nodules of *Vicia faba* have intrinsically high levels of cytokinin (Henson and Wheeler, 1976). The application of chlormequat (Hassan and El-Moursi, 1982), resulted in an increase in root nodulation. Therefore, this may be part of the reason for the increased pod set associated with the application of anti-gibberellins.

The aim of this experiment was to apply the plant growth regulators (at the same growth stage as already determined as being the most advantageous in Chapter 3); and to monitor the growth of the treated plants by separating the component parts in order to provide a more detailed knowledge of the effects of such chemicals on plant growth and provide some evidence to support the above hypothesis

Method

Two seeds of the variety Threefold White were sown into 15cm pots containing Levington potting compost and perlite during January 1989 in a bee-proof glasshouse at the Bush Estate, Edinburgh School of Agriculture. Seedlings were singled and staked to prevent lodging. The plants were subjected to minimum day-time and night-time temperatures of 21 and 16°C respectively. Supplementary

lighting was provided by 400W sodium lamps suspended 1m above the plants, to achieve a 16 hour photoperiod.

The experiment was arranged as a randomised block, each treatment was replicated 5 times. All treatments were applied at growth stage 09 with the aid of a hand-held sprayer until run-off. Treatments were: Control (Water + "Agral" wetter); Alar, (1g/l + wetter); JF 10405, (2ml/l + wetter) and EL500 (0.25g/l + wetter). All flowers were hand-tripped to ensure pollination.

Four separate measurement times were used in order obtain a clear indication of plant development throughout the growing period; growth stages 41, 91, the end of flowering and final harvest. At all timings plant height was recorded, then mainstems were cut off at soil level and the plants divided up into their component parts of roots, stem and leaves. Pod set was measured on all plants as a check to ensure all measurement times were comparable and that the plants had received the correct growth regulator application. In addition to this, at growth stage 91 and the end of flowering, the pods were weighed. At final harvest, the plants were also scored for distribution of harvestable pods, weight of pods, weight and number of beans and the weight of root nodules.

As the plants were grown in a mixture of compost and perlite, the separation of the roots from the soil was much easier. Separation was achieved by careful rinsing of the roots over a set of graduated sieves with particles of compost and perlite being removed with forceps, so ensuring all root parts were collected and recorded. At harvest root nodules were also separated with the use of forceps. All components of the plants were weighed at the time of separation, then placed directly into individual polythene bags and frozen until the dry weights could be carried out. Dry weights were achieved by placing the samples into an oven set at 90°C for as long as necessary to ensure equilibrium weight (generally 48-52 hours).

Results

Intra-Raceme Pod Set

Typical pod set distribution was displayed by control plants at growth stage 91, with average percentage pod set on the proximal three flower positions being 28%.

Average percentage pod set on the distal four positions was 4%. The application of Alar, increased the average percentage on the first three flower positions to 31%, while pod set on the distal four flowers was increased to 6% (Fig.5.1a). JF 10405 increased average pod set on the three proximal flowers to 52.5%, however, these increases were not significant at any individual site. Pod set on the distal flowers was increased to 11%. EL500 increased pod set at flower position 3 ($p < 0.01$) to 35%, so causing average pod set at the three most proximal flowers to be increased to 58% (Fig. 5.1a). Average pod set on the distal four flowers was 7%.

Control plants measured at the end of flowering exhibited slightly increased levels of pod set at the proximal 3 flowers compared to those plants measured at growth stage 91, with the average being 31%. Average pod set on the distal four flowers was 8%. The application of Alar increased the average pod set of the three proximal flowers to 39%, while distal flower pod set was reduced to 3% (Fig. 5.2a). Levels of pod set on the three proximal flowers treated with JF 10405, were similar to those of the Alar treated plants i.e. 40%. Average pod set at the distal flowers was 16%. The application of EL500, resulted in similar levels of pod set to control plants at the first three flower sites. Pod set on the distal four flowers was 6%.

Percentage pod set on the plants that were measured at harvest showed no significant increases at any individual flower positions compared to control plants, which had high levels of pod set, particularly at the proximal two flower positions (Fig. 5.3a).

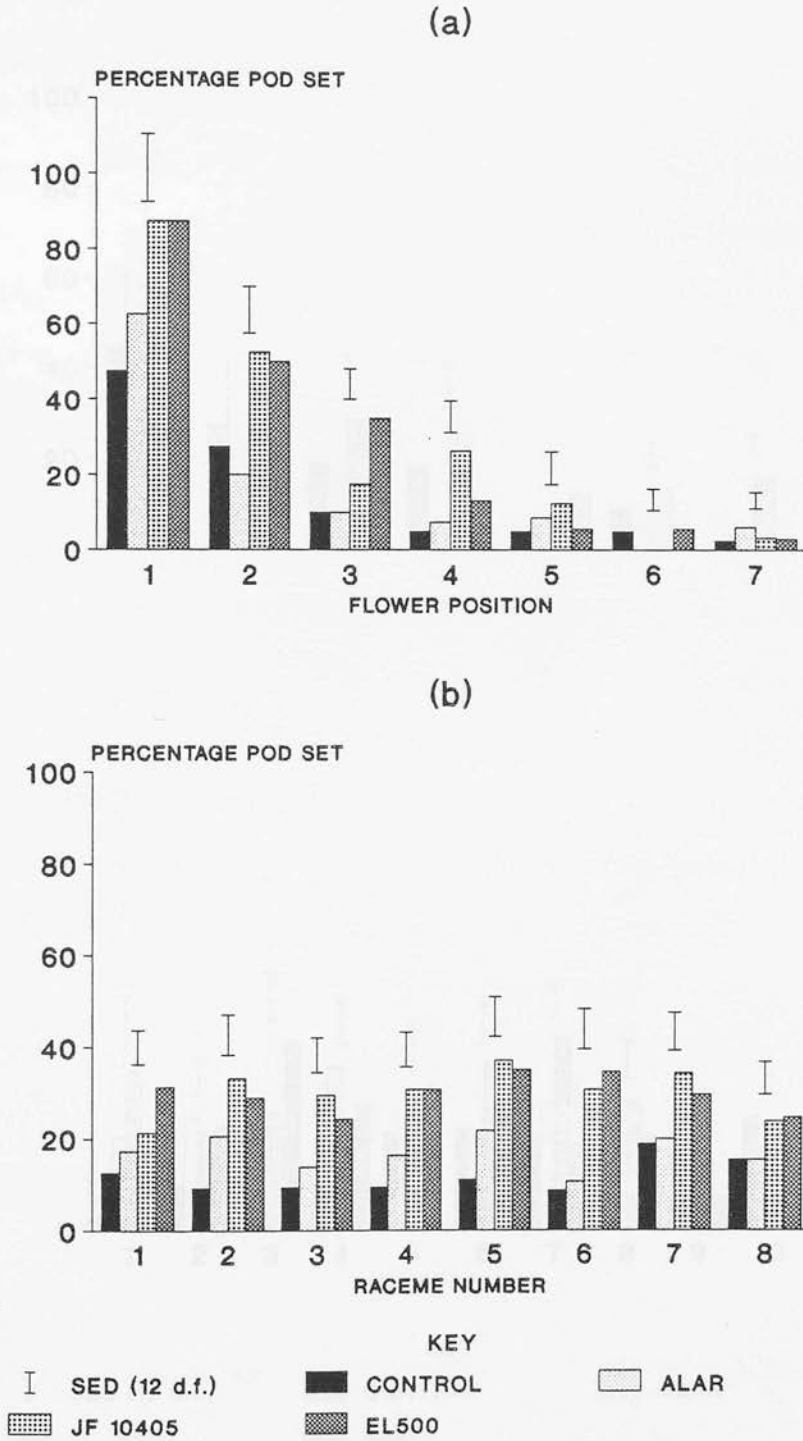


Figure 5.1: Effect of Alar, JF 10405 and EL500 on a) intra-raceme and b) inter-raceme pod set, at growth stage 91. (Actual figures are shown in Appendices 5.1, 5.2)

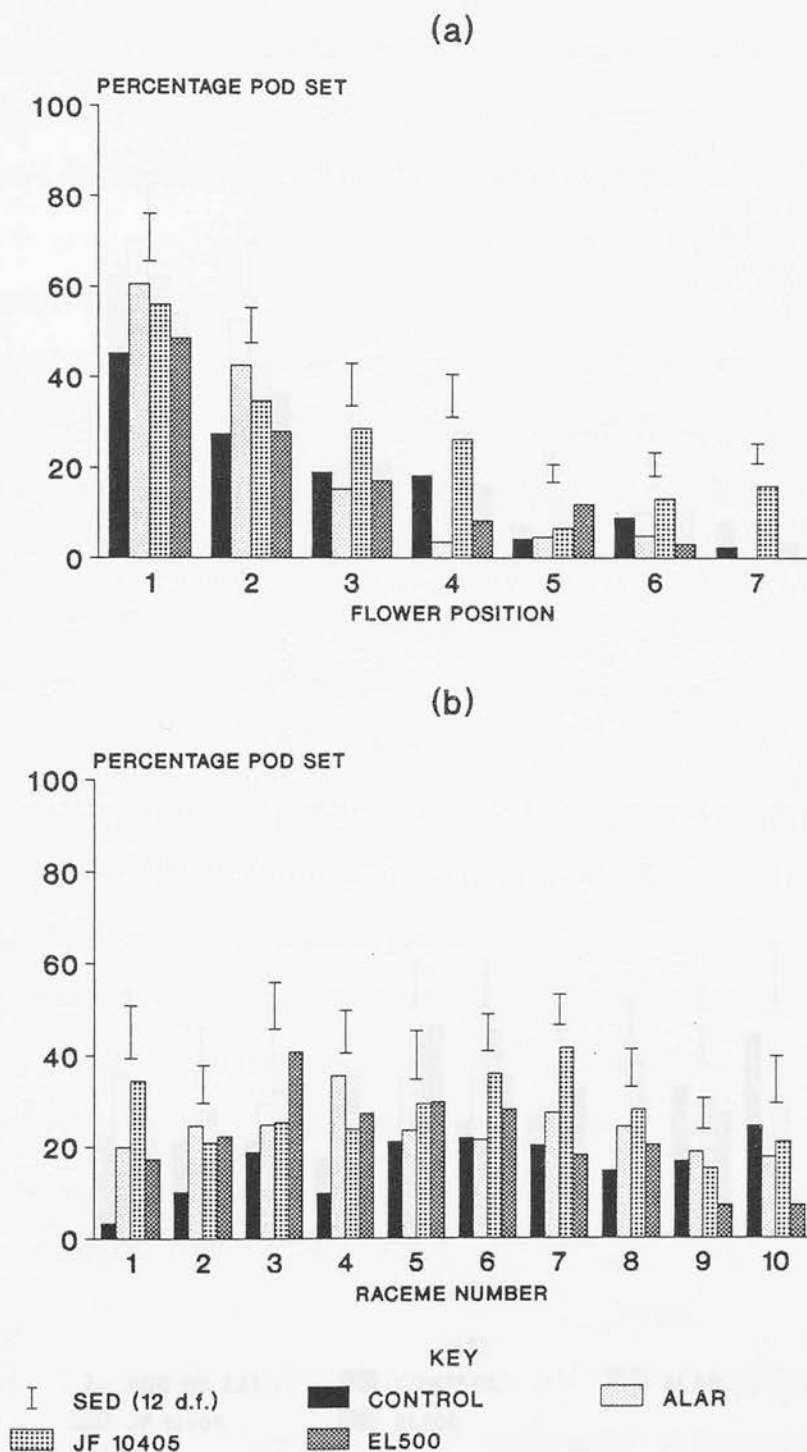
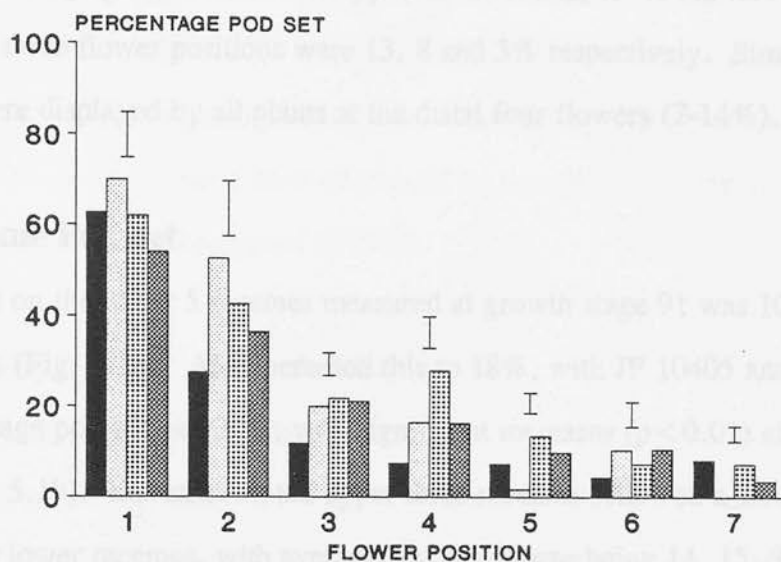
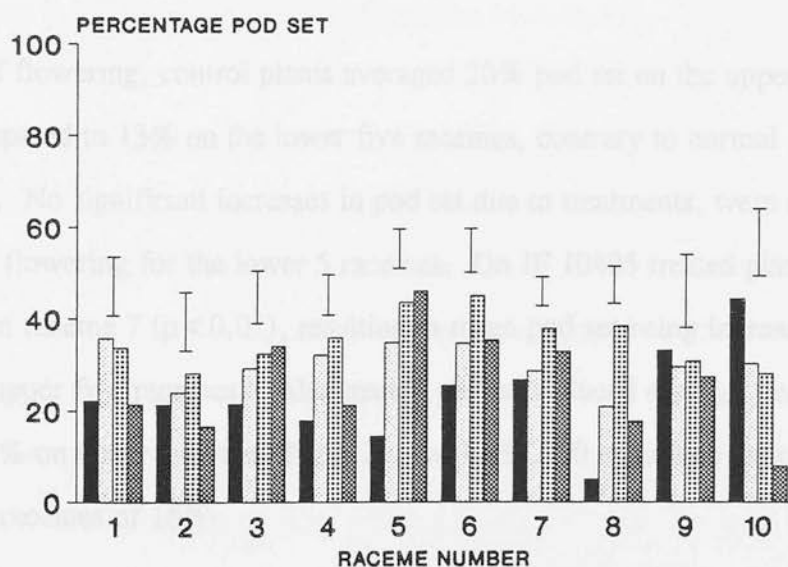


Figure 5.2: Effect of Alar, JF 10405 and EL500 on a) intra-raceme and b) inter-raceme pod set at the end of flowering. (Actual figures are shown in Appendices 5.3, 5.4)

(a)



(b)



KEY

I SED (12 d.f.)

■ CONTROL

□ ALAR

▤ JF 10405

▨ EL500

Figure 5.3: Effect of Alar, JF 10405 and EL500 on a) intra-raceme and b) inter-raceme pod set measured at harvest. (Actual figures are shown in Appendices 5.5, 5.6)

Increases in average pod set due to the application of Alar, JF 10405 and EL500, at the proximal three flower positions were 13, 8 and 3% respectively. Similar levels of pod set were displayed by all plants at the distal four flowers (7-14%).

Inter-Raceme Pod Set

Mean pod set on the lower 5 racemes measured at growth stage 91 was 10% in control plants (Fig. 5.1b). Alar increased this to 18%, with JF 10405 and EL500 mean percentage pod set was 30%, with significant increases ($p < 0.01$) at racemes 4 and 5 (Fig. 5.1b). Increases on the upper three racemes followed a similar pattern to the lower racemes, with average pod set figures being 14, 15, 30 and 30% respectively.

At the end of flowering, control plants averaged 20% pod set on the upper five racemes compared to 13% on the lower five racemes, contrary to normal observations. No significant increases in pod set due to treatments, were recorded at the end of flowering for the lower 5 racemes. On JF 10405 treated plants, 41% of pods set on raceme 7 ($p < 0.01$), resulting in mean pod set being increased to 28% on the upper five racemes. Alar treated plants produced average pod set figures of 22% on upper racemes (Fig 5.2b), while EL500 caused an average pod set on upper racemes of 16%.

On control plants measured at harvest, the pattern of pod set exhibited was similar to that measured at the end of flowering i.e. pod set increased on the upper five racemes to 27% compared to 19% on the lower five racemes (Fig. 5.3b). As at the end of flowering, JF 10405 treatments resulted in levels of pod set that were quite

uniform over all racemes. A 32% increase in pod set ($p < 0.01$) at raceme 5, resulted in the plants treated with EL500 having an increased average pod set figure on the lower five racemes of 28%. This was, however, lower than the average exhibited by the other two treatments, due to a lesser increase at other racemes. Only JF 10405 caused the mean level of pod set on the upper racemes to increase above control plants (36% compared to 27%).

Intra-Raceme Percentage Harvestable Pods

The pattern of percentage harvestable pods on all plants displayed the typical pattern of proximal dominance (Fig. 5.4a). In control plants almost all of the harvestable pods were sited at flower position 1. No significant increases at any flower positions were evident due to any treatment, however, increases were seen at flower position 2 from 8.3% in control plants to between 11.3-12.2%.

Inter-Raceme Percentage Harvestable Pods

Control plants had similar levels of percentage harvestable pods at all racemes, the average being 9% at both the lower 5 and upper 5 racemes (Fig. 5.4b). The application of Alar increased the number of harvestable pods at all racemes except raceme 10, but these increases were not significant. JF 10405 treated plants possessed fewer harvestable pods at raceme 1, but due to a significant increase ($p < 0.01$) at raceme 5, average pod set on the lower 5 racemes increased to 12%. The application of EL500 resulted in no significant increases in the percentage of harvestable pods at any raceme.

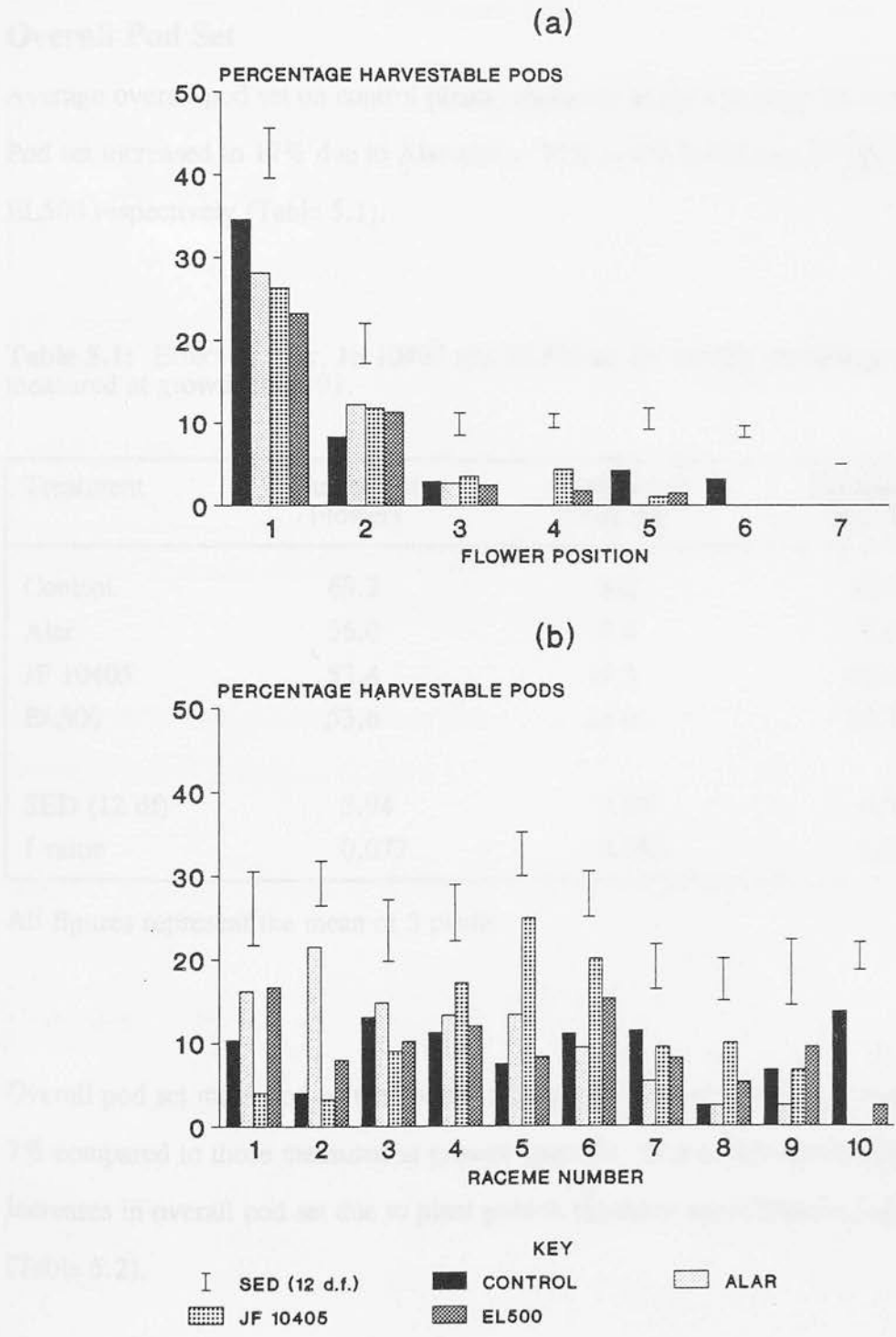


Figure 5.4: Effect of Alar, JF 10405 and EL500 on a) intra-raceme and b) inter-raceme distribution of harvestable pods. (Actual figures are shown in Appendices 5.7, 5.8)

Overall Pod Set

Average overall pod set on control plants, measured at growth stage 91 was 12%. Pod set increased to 17% due to Alar and to 30% ($p < 0.01$) due to JF 10405 and EL500 respectively (Table 5.1).

Table 5.1: Effect of Alar, JF 10405 and EL500 on the overall percentage pod set, measured at growth stage 91.

Treatment	Number of Flowers	Number of Pods Set	Percentage Pod Set
Control	63.2	8.2	12.0
Alar	56.0	9.4	17.0
JF 10405	52.4	15.6	30.0
EL500	53.6	16.0	30.0
SED (12 df)	3.94	2.90	4.50
f value	0.072	0.036	0.003

All figures represent the mean of 5 plants

Overall pod set measured on the control plants at the end of flowering, increased by 7% compared to those measured at growth stage 91. Due to this no significant increases in overall pod set due to plant growth regulator application were observed (Table 5.2).

Table 5.2: Effect of Alar, JF 10405 and EL500 on overall pod set, measured at the end of flowering.

Treatment	Number of Flowers	Number of Pods Set	Percentage Pod Set
Control	108.2	19.4	19.0
Alar	104.4	21.2	20.0
JF 10405	109.8	28.6	26.0
EL500	101.0	18.6	19.0
SED (12 df)	15.86	4.41	4.70
f value	0.944	0.148	0.351

All figures represent the mean of 5 plants

Overall percentage pod set on those plants scored at harvest, showed no significant increases due to treatments (Table 5.3).

Table 5.3: Effect of Alar, JF 10405 and EL500 on overall percentage pod set, measured at harvest.

Treatment	Number of Flowers	Number of Pods Set	Percentage Pod Set	Number of Harvestable Pods	Percentage Harvestable Pods
Control	93.2	17.4	21.0	7.4	8.5
Alar	101.6	26.4	26.0	7.0	7.0
JF 10405	110.4	30.6	28.0	8.0	7.6
EL500	105.8	23.2	23.0	6.6	6.5
SED (12 df)	19.98	4.25	4.35	1.26	1.52
f value	0.847	0.052	0.319	0.724	0.603

All figures represent mean of 5 plants

Overall Percentage Harvestable Pods

The application of Alar, JF 10405 and EL500 decreased the overall percentage of harvestable pods compared to the control plants by between 0.9 - 2.0% (Table 5.3). These effects were not significant.

Mainstem Yield

Number of harvestable pods was reduced by the application of Alar and EL500, but increased by the application of JF 10405, these results were not significant. Fresh weight of beans was however increased by Alar (7%), JF 10405 (8%) and decreased by 11 % with EL500. Dry weight of beans followed a similar pattern. Once again these effects were not significant (Table 5.4).

Table 5.4: Effect of Alar, JF 10405 and EL500 on mainstem yield

Treatment	Number of Harvestable Pods	Fresh Weight of Harvestable Pods (g)	Fresh Weight of Beans (g)	Dry Weight of Beans (g)
Control	7.4	138.6	53.5	17.5
Alar	7.0	128.2	57.7	21.9
JF 10405	8.0	149.4	58.3	17.3
EL500	6.6	126.8	48.0	15.3
SED (12 df)	1.26	25.70	12.29	4.41
f value	0.724	0.801	0.824	0.519

All figures represent the mean of 5 plants

Plant Height

Vegetative height did not alter due to treatments or between timings (Figure 5.5).

Reproductive height was reduced by on average 25% at all timings, by applications of Alar, JF 10405 and EL500.

Control plants were 78cm taller at final harvest than at growth stage 41. All treated plants grew until the end of flowering stage, then little difference was seen in reproductive growth between that stage and the final harvest. Control plants, however, continued to grow by 10% during this period.

Distribution of Dry Matter

Growth Stage 41

Dry matter distribution was divided into stem, root and leaves, as no substantial development of pods had occurred (Fig. 5.6). Control plants had 44% of dry matter in the stem. Although plant height was reduced by the application of all treatments no significant difference in the percentage of dry matter contained in the stems of treated plants was observed. The percentage of dry matter contained within the leaves was decreased by Alar and JF 10405 to 32.5% and 30.8% respectively compared to control plants (33.3%), whereas EL500 caused an increase to 35.5%. None of these effects were significant. All treatments caused more dry matter to be contained in the roots, with increases over control plants being; Alar (1.1%), JF 10405 (1.6%) and EL500(1.2%). These effects were again not significant.

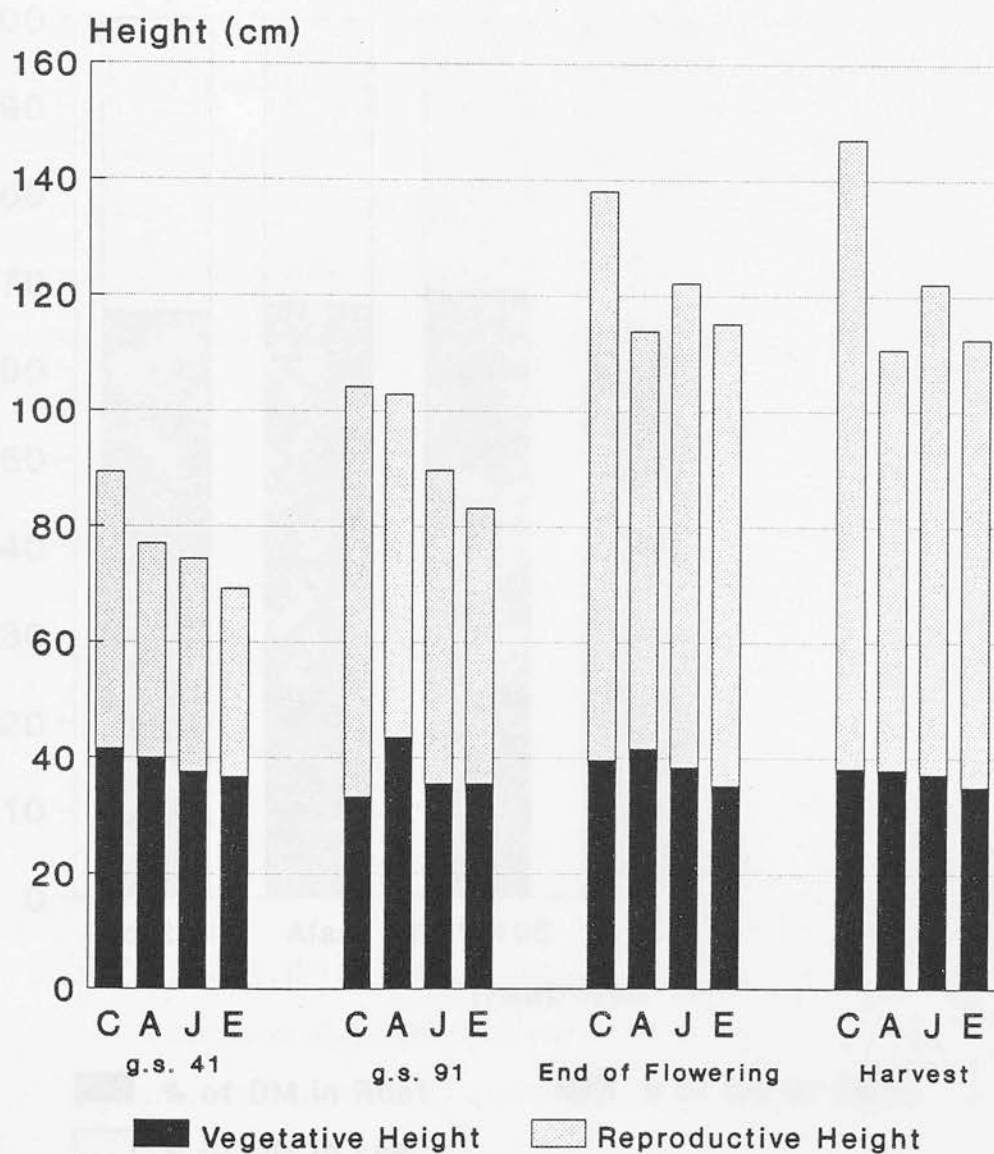


Figure 5.5: Effect of Alar (A), JF 10405 (J) and EL500 (E) on vegetative and reproductive height, measured at growth stages 41, 91, end of flowering and final harvest compared to control plants (C). (Actual figures are shown in Appendices 5.12, 5.13, 5.14)

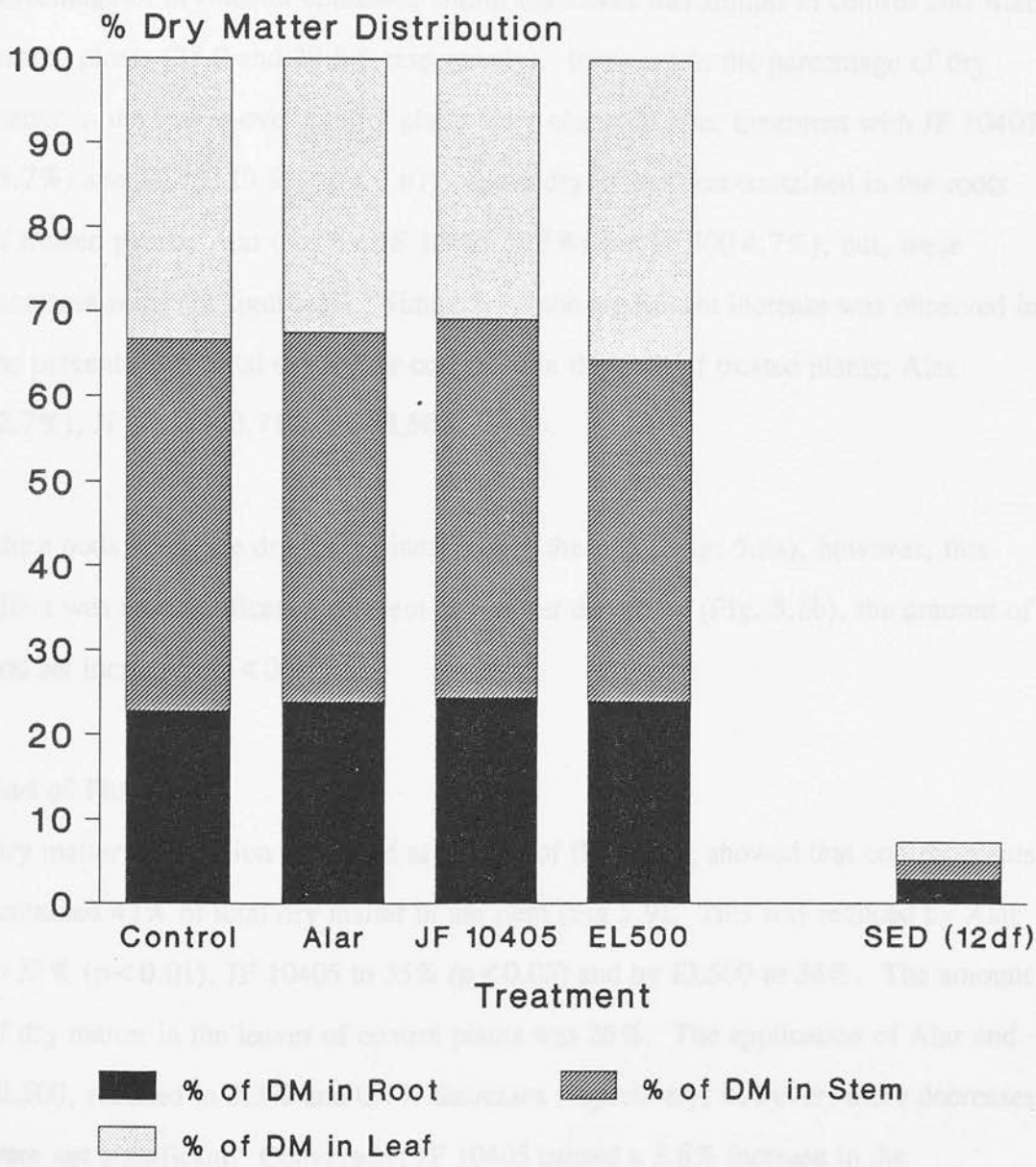


Figure 5.6: Effect of Alar, JF 10405 and EL500 on the distribution of dry matter, at growth stage 41. (Actual figures are shown in Appendix 5.9)

Growth Stage 91

The percentage of total dry matter contained in the stem was reduced between 8.9 - 10.2% ($p < 0.001$) by all treatments compared to the control (Fig. 5.7). The percentage of dry matter contained within the leaves was similar in control and Alar treated plants (28.0 and 28.8% respectively). Increases in the percentage of dry matter in the leaves over control plants were observed after treatment with JF 10405 (3.7%) and EL500 (9.9%, $p < 0.01$). More dry matter was contained in the roots of treated plants; Alar (5.5%), JF 10405 (4.5%) and EL500 (4.7%), but, these increases were not significant. Similarly, a non-significant increase was observed in the percentage of total dry matter contained in the pods of treated plants; Alar (2.7%), JF 10405 (0.7%) and EL500 (3.3%).

More pods set as the dry matter increased in the roots (Fig. 5.8a), however, this effect was not significant. As stem dry matter decreased (Fig. 5.8b), the amount of pod set increased ($p < 0.05$).

End of Flowering

Dry matter distribution measured at the end of flowering, showed that control plants contained 43% of total dry matter in the stem (Fig 5.9). This was reduced by Alar to 32% ($p < 0.01$), JF 10405 to 35% ($p < 0.05$) and by EL500 to 36%. The amount of dry matter in the leaves of control plants was 26%. The application of Alar and EL500, resulted in 3.3% and 0.4% decreases respectively, however, these decreases were not significant. Conversely, JF 10405 caused a 3.8% increase in the percentage of dry matter contained within the leaves. All treatments resulted in an increase in the amount of dry matter contained within the roots; Alar, 1.6%; JF 10405, 2.0% and EL500, 1.6%, all non-significant. Due to pod set being enhanced by plant growth regulator treatments, the percentage of total dry matter contained within the pods was increased compared to control plants. With Alar it was 12.4% ($p < 0.01$), with EL500, 5.4% and JF 10405, 2.0%.

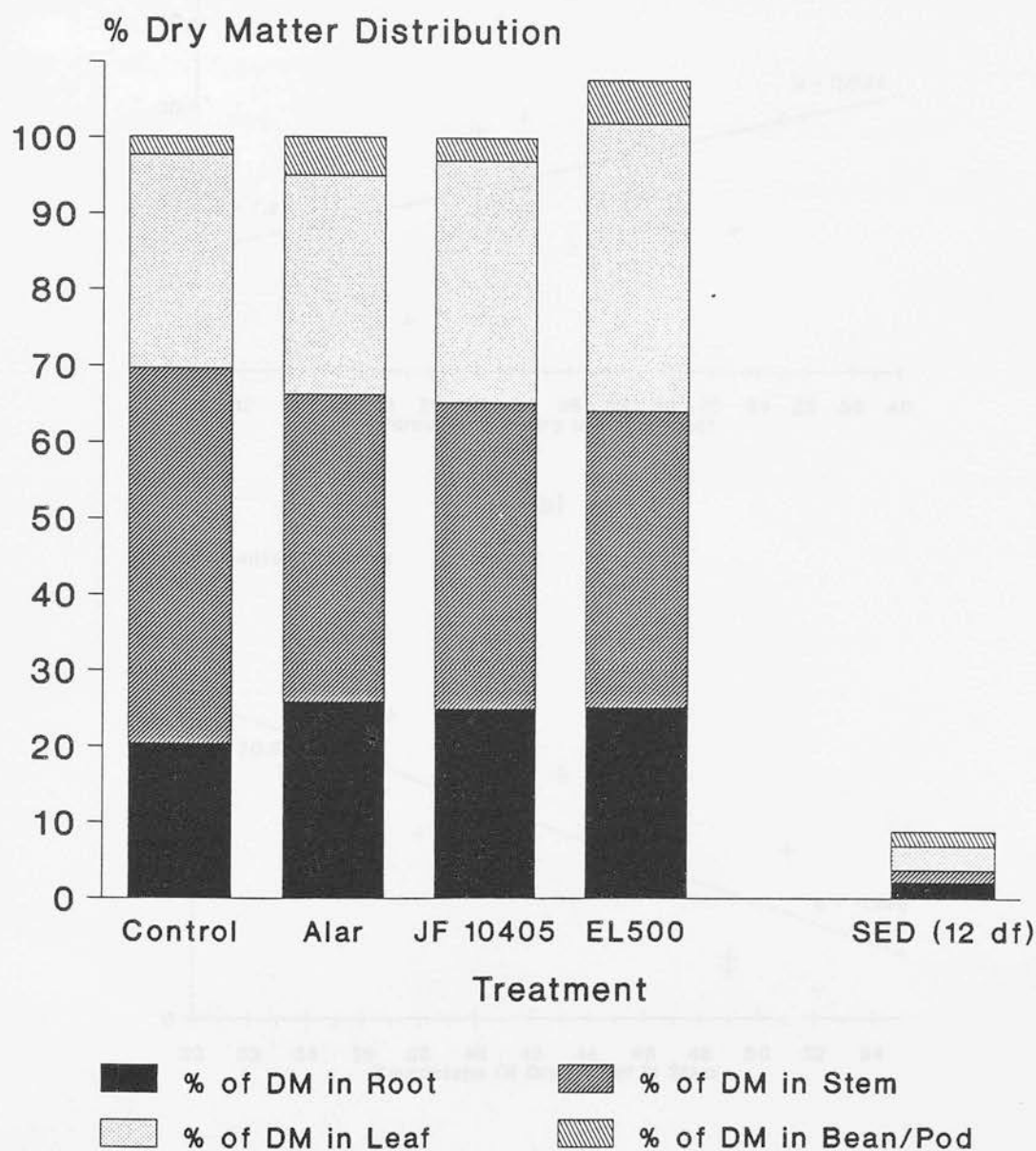


Figure 5.7: Effect of Alar, JF 10405 and EL500 on the distribution of dry matter at growth stage 91. (Actual figures are contained in Appendix 5.10).

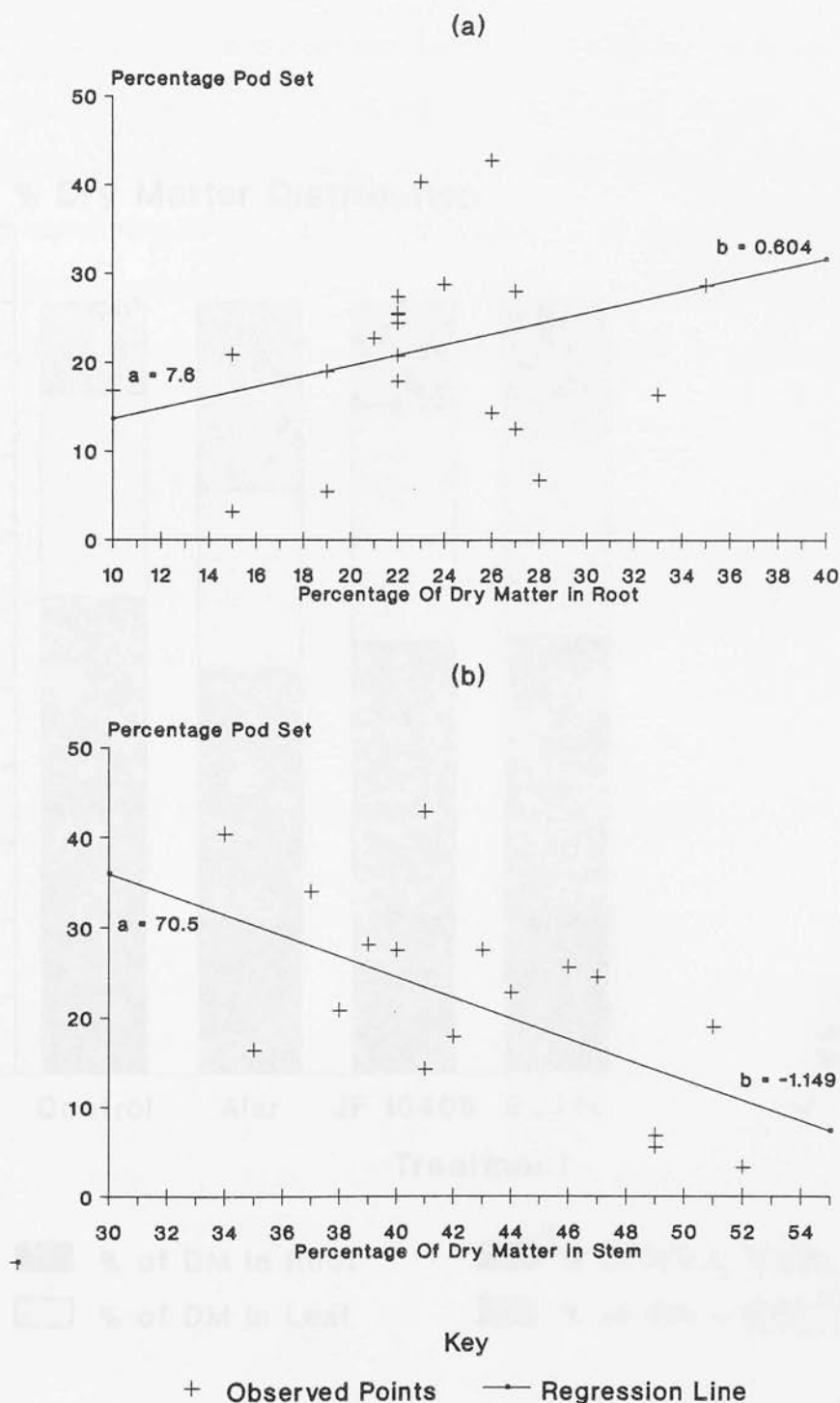


Figure 5.8: Relationship between a) percentage of dry matter contained in the root and percentage pod set and b) percentage of dry matter contained in the stem and percentage pod set, at growth stage 91.

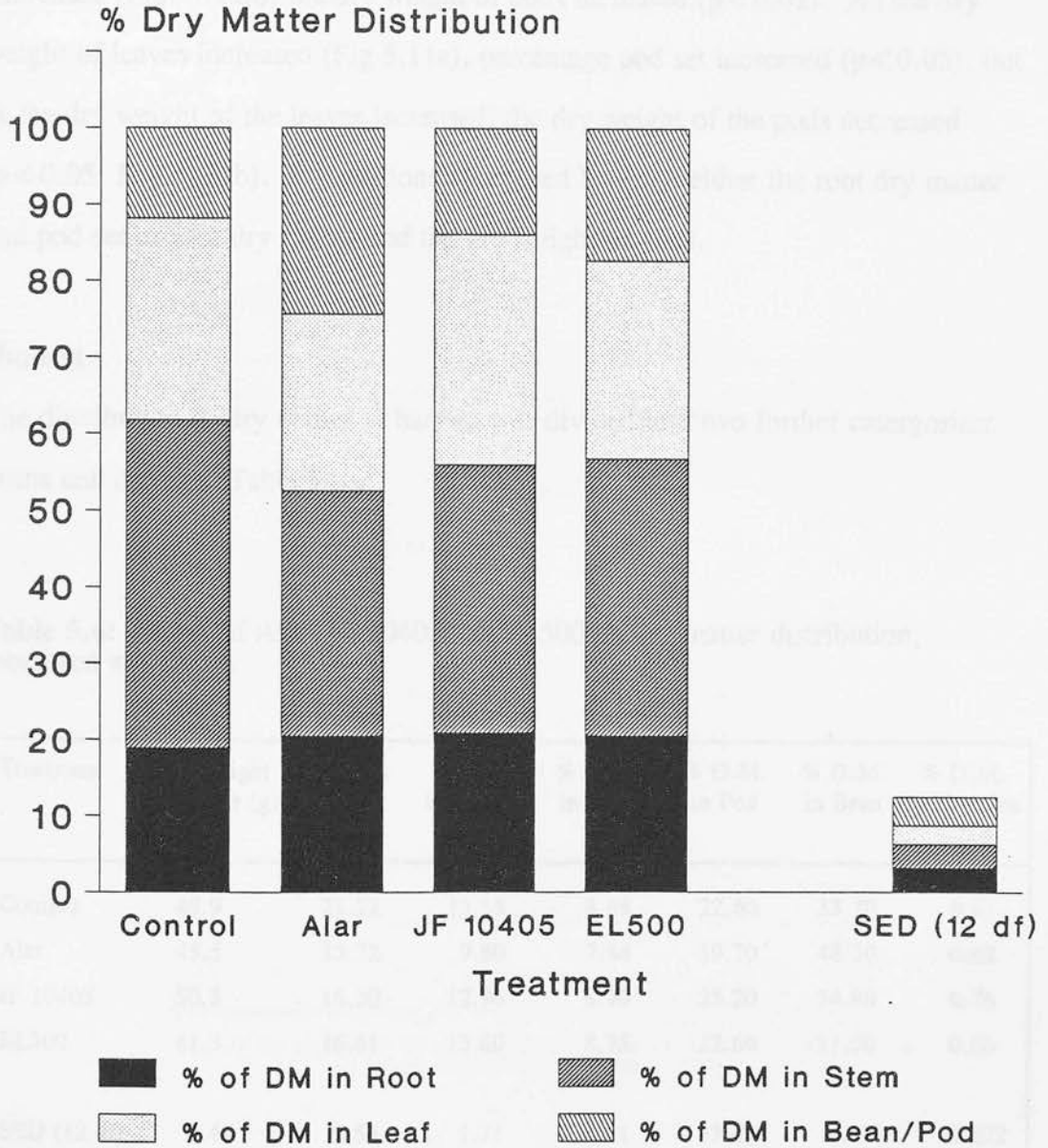


Figure 5.9: Effect of Alar, JF 10405 and EL500 on the distribution of dry matter at the end of flowering. (Actual figures are shown in Appendix 5.11)

No definite relationship existed between the amount of total dry matter contained within the stem and percentage pod set (Fig. 5.10a), however, as stem dry weight decreased (Fig. 5.10b), the dry weight of pods increased ($p < 0.01$). As the dry weight of leaves increased (Fig 5.11a), percentage pod set increased ($p < 0.05$), but as the dry weight of the leaves increased, the dry weight of the pods decreased ($p < 0.05$, Fig. 5.11b). No relationship existed between either the root dry matter and pod set or root dry matter and the dry weight of pods.

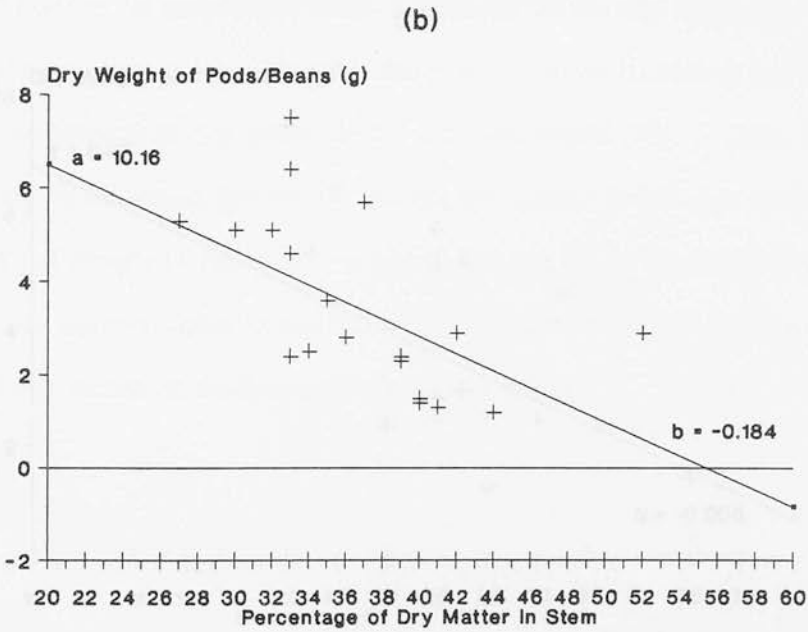
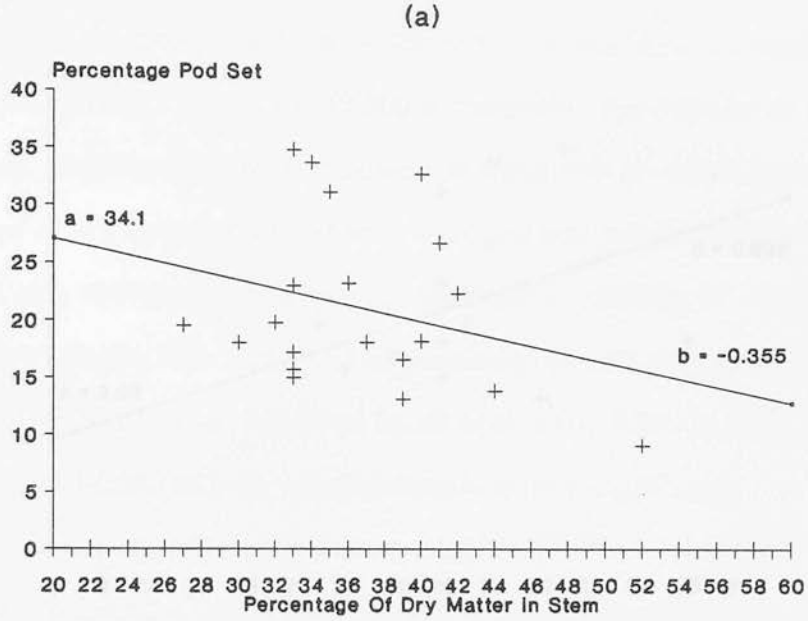
Harvest

The distribution of dry matter at harvest was divided into two further categories: beans and nodules (Table 5.4).

Table 5.4: Effect of Alar, JF 10405 and EL500 on dry matter distribution, measured at harvest.

Treatment	Total Weight Dry Matter (g)	% D.M. in Stem	% D.M. in Leaves	% D.M. in Root	% D.M. in Pod	% D.M. in Bean	% D.M. in Nodules
Control	49.9	21.22	13.18	8.48	22.60	33.70	0.81
Alar	45.5	13.72	9.80	7.88	19.70	48.30	0.62
JF 10405	50.3	16.30	12.96	9.90	25.20	34.90	0.76
EL500	41.3	16.61	13.60	8.75	22.60	37.90	0.56
SED (12 df)	8.43	2.54	1.71	1.31	3.03	6.13	0.272
f value	0.687	0.072	0.156	0.500	0.388	0.124	0.762

All figures represent the mean of 5 plants



Key

+ Observed Points — Regression Line

Figure 5.10: Relationship between a) percentage of dry matter contained in the stem and percentage pod set and b) percentage of dry matter contained in the stem and dry weight of pods, at the end of flowering

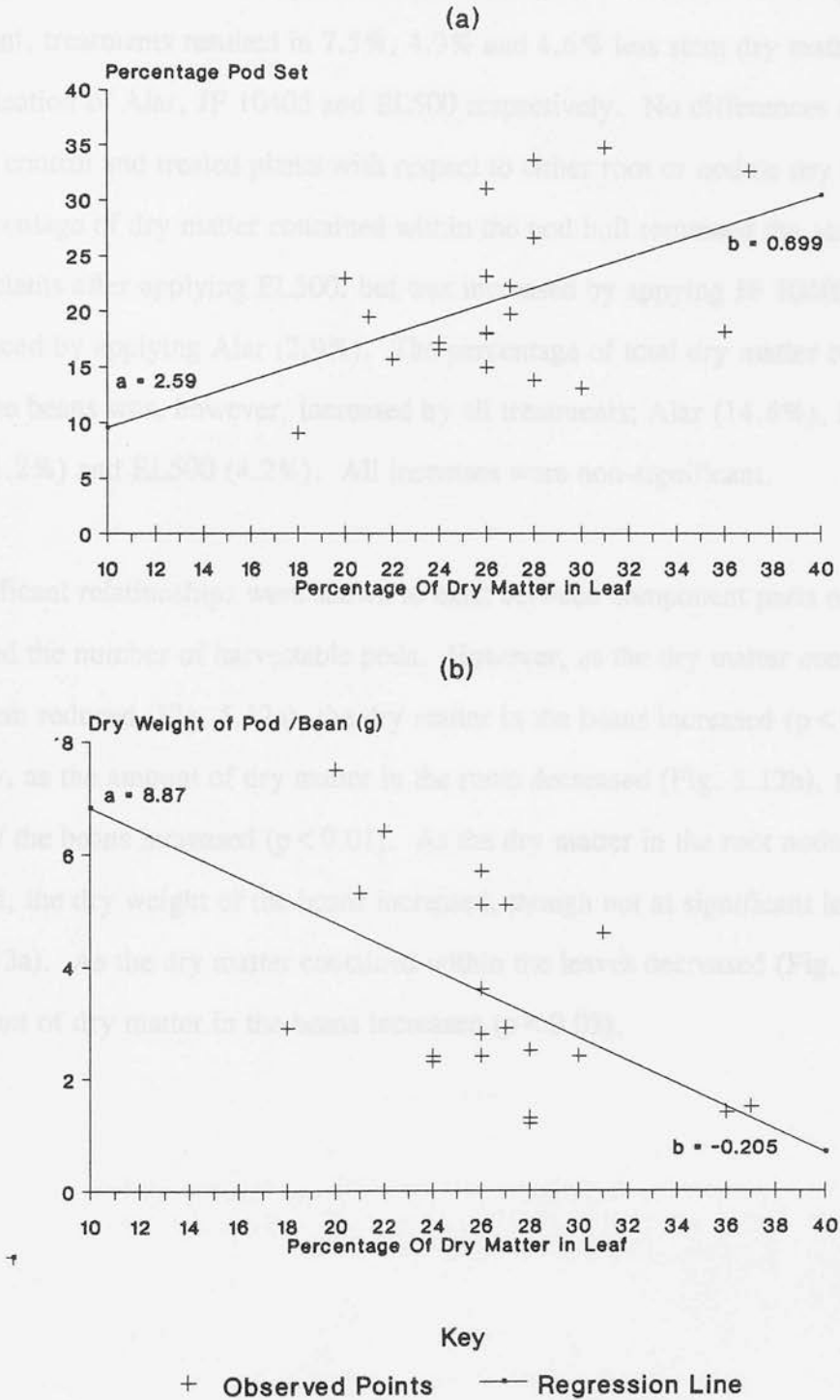


Figure 5.11: Relationship between a) percentage of dry matter contained in the leaves and percentage pod set and b) percentage of dry matter contained in the leaves and dry weight of pods, measured at the end of flowering.

Control plants had 21.2% of dry matter contained within the stem. Although non-significant, treatments resulted in 7.5%, 4.9% and 4.6% less stem dry matter, with the application of Alar, JF 10405 and EL500 respectively. No differences occurred between control and treated plants with respect to either root or nodule dry matter. The percentage of dry matter contained within the pod hull remained the same as in control plants after applying EL500, but was increased by applying JF 10405 (2.6%) and reduced by applying Alar (2.9%). The percentage of total dry matter contained within the beans was, however, increased by all treatments; Alar (14.6%), JF 10405 (1.2%) and EL500 (4.2%). All increases were non-significant.

No significant relationships were shown to exist between component parts of the plants and the number of harvestable pods. However, as the dry matter contained in the stem reduced (Fig. 5.12a), the dry matter in the beans increased ($p < 0.05$). Similarly, as the amount of dry matter in the roots decreased (Fig. 5.12b), the dry matter of the beans increased ($p < 0.01$). As the dry matter in the root nodules increased, the dry weight of the beans increased, though not at significant levels (Fig. 5.13a). As the dry matter contained within the leaves decreased (Fig. 5.13b), the amount of dry matter in the beans increased ($p < 0.05$).

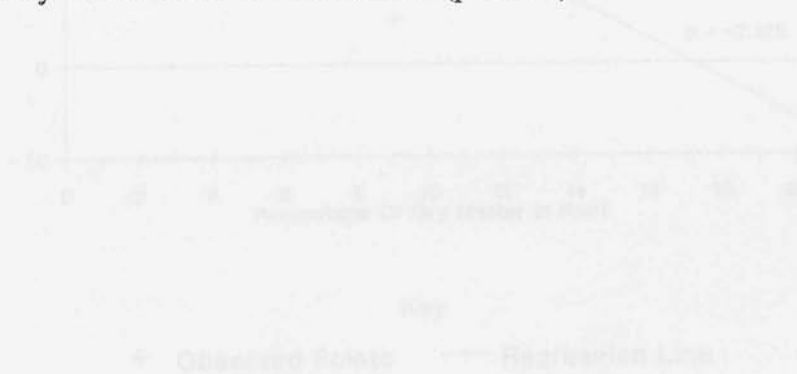
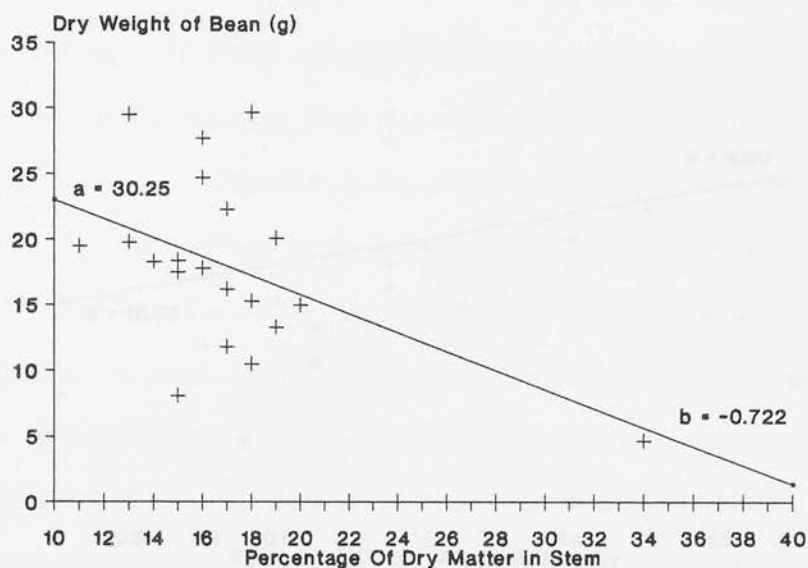
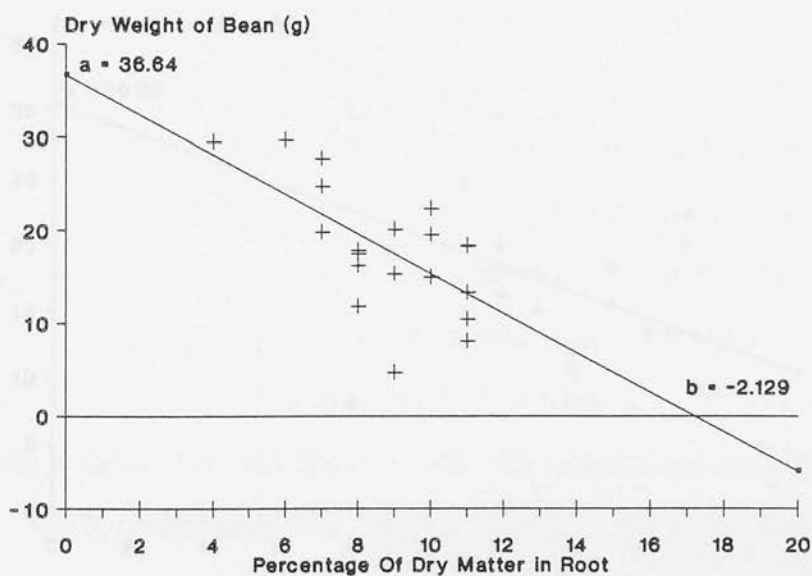


Figure 5.12a Relationship between a) percentage of dry matter contained in the stem and dry weight of beans and b) percentage of dry matter contained in the roots and dry weight of beans, measured at harvest.

(a)



(b)



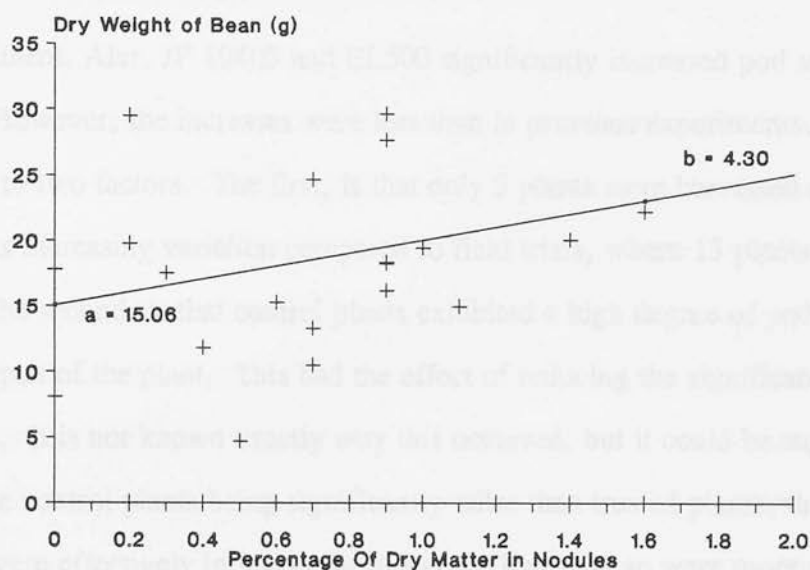
Key

+ Observed Points —●— Regression Line

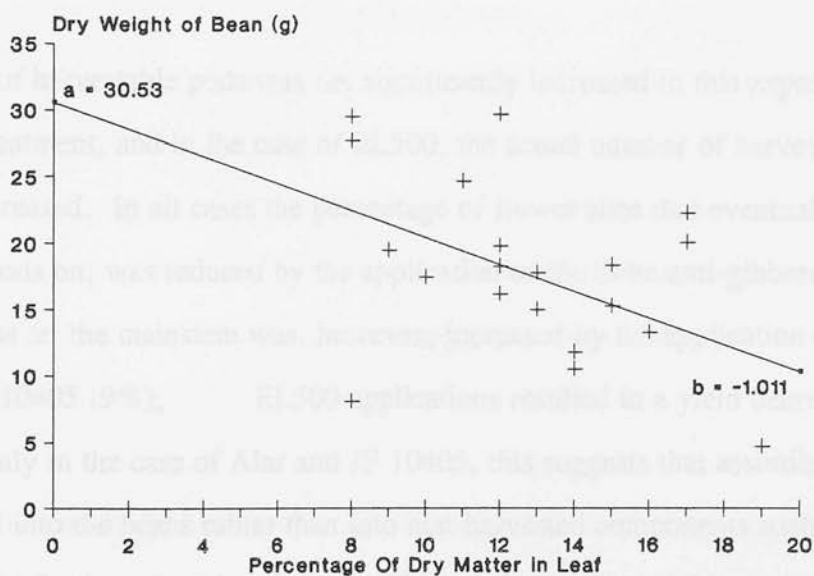
Figure 5.12: Relationship between a) percentage of dry matter contained in the stem and dry weight of beans and b) percentage of dry matter contained in the roots and dry weight of beans, measured at harvest.

Discussion

(a)



(b)



Key

+ Observed Points — Regression Line

Figure 5.13: Relationship between a) percentage of dry matter contained in the nodules and dry weight of beans and b) percentage of dry matter contained in the leaves and dry weight of beans, measured at harvest.

Discussion

In this experiment, Alar, JF 10405 and EL500 significantly increased pod set in *Vicia faba*. However, the increases were less than in previous experiments. This could be due to two factors. The first, is that only 5 plants were harvested at any one time, thus increasing variation compared to field trials, where 15 plants were harvested. The second, is that control plants exhibited a high degree of pod set towards the apex of the plant. This had the effect of reducing the significance of any increases. It is not known exactly why this occurred, but it could be suggested that due to the control plants being significantly taller than treated plants, the tops of these plants were effectively in less competition for light and so were more able to set pods.

The number of harvestable pods was not significantly increased in this experiment due to any treatment, and in the case of EL500, the actual number of harvestable pods was decreased. In all cases the percentage of flower sites that eventually had harvestable pods on, was reduced by the application of the three anti-gibberellins. Yield of beans on the mainstem was however, increased by the application of Alar (8%) and JF 10405 (9%). EL500 applications resulted in a yield decrease of 11%. Certainly in the case of Alar and JF 10405, this suggests that assimilates were directed into the beans rather than into non-harvested components such as the stem. The distribution of total dry matter (Table 5.4), confirmed this, as treated plants contained a greater percentage of the total dry matter in the beans.

At growth stage 91, the application of growth regulators increased pod set by between 4.8 and 18%. The amount of dry matter contained within the stem was reduced significantly by the application of the growth regulators, while dry matter increased in roots and leaves. Attiya *et al.* (1983), suggested that the increase in pod set associated with the application of paclobutrazol was due to a diversion of

assimilates from vegetative growth to reproductive sinks. The main vegetative sink is the stem. Results from this experiment support this supposition.

It was shown in previous experiments that GA_3 applied to flowers 24 hours before tripping had the effect of decreasing pod set on lower racemes, while applications made post tripping had little effect (Rylott and Smith, 1990). The application of cytokinin (as BAP), increased overall pod set by 85 and 78% at pre and post tripping applications. Thus it can be suggested, that to ensure pod set on the lower racemes of the plant, endogenous hormones must be in a ratio of high levels of cytokinin and low levels of gibberellin. It could also be suggested that the observed increase in the percentage of total dry matter in the roots (Fig. 5.7), combined with a decrease in stem length and dry matter contained within it, caused by the application of growth regulators, increased the available cytokinin:gibberellin ratio. This change in the ratio of endogenous hormones then favoured increased pod set, especially at lower racemes (Fig. 5.1). The increase in the amount of dry matter contained within the leaves also aided pod set (Fig. 5.11a), as the assimilates produced by the leaves were diverted more directly into the developing reproductive sinks, where the increased cytokinin levels promote vascular differentiation and importation of assimilates.

A positive relationship was observed at the end of flowering between the percentage of total of dry matter contained in the leaves and percentage pod set (Fig. 5.11a). JF 10405 was the only treatment to increase the number of pods set at this stage and was also the only treatment to increase the percentage of total dry matter contained in the leaves. This, in conjunction with Fig. 5.11a, suggests that assimilates produced by the leaves remains a major factor in pod set and pod development until the end of flowering.

Numbers of pods set per plant at the end of flowering was similar for both control and treated plants. The percentage of dry matter contained within these pods, however, increased compared to control plants (Fig. 5.9). This suggests growth regulators applied at growth stage 09, have the effect of continuing to cause a change in the balance of endogenous plant growth substances, so allowing assimilates to be more available to the developing reproductive sinks. At the end of flowering, treated plants had on average 8.4% less total dry matter contained in the stem, while 1.7% more was contained within the roots. Thus, treated plants may continue to have a higher cytokinin:gibberellin ratio compared to control plants. Cytokinins attract assimilates especially to developing reproductive sinks (due to cell division and hence increased sink capacity), while gibberellins appear to favour growth of the most dominant sink (the stem) and this may explain why the treated plants had a greater percentage of total dry matter contained within the pods.

The period between the end of flowering and harvest is primarily when the pods "fill". The average number of harvestable pods on all plants from all treatments at harvest was similar, ranging from 7.4 in control plants to between 6.6 and 8.0 in treated plants. The fresh weight of beans, however, was increased over control plants in Alar and JF 10405 treatments and was reduced in EL500 treatments. Beans harvested from treated plants contained a greater percentage of the total dry matter of the plant than control plants and this therefore increased their harvest indices.

Part of the reason for this, is the continued effect of the growth regulators upon the growth of the stem. Indeed, Ismail and Sagar (1980), found that fed leaves of broad beans failed to export more than 40% of the ^{14}C that they initially fixed during the first 24 hours and that the stem was the principal sink for the radio active carbon. Control plants grew by 10% after the end of flowering, compared to treated plants, that on average stopped growing. Thus, assimilates in control plants

were effectively wasted by adding to non-essential stem growth, which remained as the dominant sink. In treated plants, the anti-gibberellins applied at growth stage 09, had the continued effect of reducing the stem's sink capacity and so allowed assimilates to be diverted directly into the beans. In general, as the amount of total dry matter contained within the stem decreased, the dry weight of beans increased (Fig. 5.12a) and this effect was promoted by the application of plant growth regulators.

At harvest, it was found that as the proportion of total dry matter contained within the roots increased, the weight of beans decreased (Fig. 5.12b). This suggests that during "pod-fill" assimilates directed towards root growth are in immediate competition with the developing beans. Hassan and El-Moursi (1982), found that the application of chlormequat increased the number of nodules on *Vicia faba*. In this experiment applications of Alar and EL500 reduced the actual dry weight of nodules by 34 and 43% respectively while JF 10405 increased the dry weight of nodules by 13%, all differences were non-significant. Thus, findings by the above authors can neither be confirmed or contradicted by this experiment. It is suggested that further work should be carried out, with larger replicates, so ensuring more accurate results are determined.

Although non-significant, it was suggested that as the amount of total dry matter contained within the nodules increased, the dry weight of beans decreased (Fig. 5.13a). All treatments reduced the percentage of total dry matter contained within the root nodules. Thus, it may follow that due to application of plant growth regulators, the amount of nodules formed is reduced. It is known that the amount of ATP used by root nodules in the fixation of nitrogen is costly (Roughley *et al.*, 1983). Further investigations into the effect of PGRs on nodulation and its effect on dry weight of pods may therefore lead to new proposals of combinations of artificial growth manipulators and nitrogen being used in the crop as a means of

more efficient production. It should be pointed out that measurements of the amount of nodules formed at growth stages 41 and 91 were not taken. Following the theories of Henson and Wheeler (1976), it is at these stages that the nodules would have the greatest effect in terms of their cytokinin production. It is suggested that future studies should concentrate on nodule formation throughout the growing season and more replicates should be used.

A significant negative trend was shown to exist between the percentage of dry matter contained in the leaves and dry weight of beans. Ishag (1973^b), found that up to late pod set, leaf area was closely correlated with total dry matter production. After this time, dry weight of beans was not correlated to leaf area, suggesting that a) assimilates were transferred from other parts e.g. the stem to the bean (as in wheat during grain fill, Austin *et al.*, (1977)), or b), that other parts of the plant were photosynthetic.

Kipps and Boulter (1974), state that pods are photosynthetic. However, as discussed by Chapman and Peat (1978), their effectiveness is probably limited due to their shaded axillary positions. This therefore explains why the negative relationship existed at harvest in this experiment, i.e. a reduction in leaf area between the end of flowering and harvest resulted in less shading and so allowed the pods to produce more of their individual assimilate requirements. In addition, the pods at the top of control plants were considerably less shaded and closer to the light, hence the successful increase in their size during this experiment.

In conclusion, it was shown that with the application of plant growth regulators, the proportion of the total dry matter contained within the roots was increased (Figs. 5.6, 5.7, 5.9 and Table 5.5). Due to this increase in the proportion of roots to the rest of the plant and especially the stem, the ratio of cytokinin:gibberellin may have been increased. At growth stages 41 and 91, pod set increases were most

pronounced, this also corresponded to when this ratio appeared to be at a maximum. Although the actual "effective" root area appeared to have increased, so possibly leading to increased cytokinin activity, this experiment also suggested that nodule growth may not be part of the reason for increased cytokinin activity. However, it should be stated that nodule growth at these earlier growth stages was not measured and so this theory cannot be completely ruled out.

The effect that the chemicals had on reducing stem growth appears to have continued right through the growing season, with treated plants not increasing in height after the end of flowering. This in turn led to the stem losing its dominance as a sink, allowing more assimilates to be diverted to the developing pods. In addition to this, the percentage of total dry matter contained within the leaves was reduced after the end of flowering, allowing pods to produce more of their individual requirements than on non-treated plants.

Due to the decrease in apical dominance exhibited by treated plants they may have become slightly deficient in auxin, which is required after pollination, for vascular differentiation and increased pod set and fill (Rylott and Smith, 1990). It could be suggested therefore, that a combination of anti-gibberellins and cytokinin applied at growth stage 09 to increase initial pod set, followed by an application of auxin and possibly more cytokinin to the sinks at growth stage 41, would result in increased pod set, increased sink capacity and successful pod development.

Chapter 6

Effect of EL500, BAP and CLIAA applied at different growth stages and using conventional and drop leg methods of spraying.

Introduction

The application of BAP was found to increase pod set in broad beans (Rylott and Smith, 1990). In addition, it was also shown that the application of auxin was important after fertilisation in order to allow vascular differentiation at the pedicel:peduncle junction. However, application of this product directly to the flowers appeared to result in the plants gaining height. Auxin is known to sustain proximal dominance in plants (Bandruski and Nonhebel, 1984). It can be suggested therefore, that application of this product via a conventional sprayer, may add to this apical dominance and height of the plant, because of its application directly to the apex of the plant.

Drop leg sprayers have a leg attached to the boom on which the jet is the fixed and directed upwards. This allows chemical to be directed upwards from the base of the crop and is a technique often employed to apply insecticides to Brussels Sprout crops by commercial growers.

It was suggested in Chapter 5, that a combination of anti-gibberellin and cytokinin at growth stage 09, followed by either further anti-gibberellin and cytokinin or cytokinin and auxin at growth stage 41, would aid pod retention.

The aim of this experiment was therefore to test the above theory and to also assess the relevance of the drop leg method of application.

Method

Two seeds of variety Threefold White were sown into 15cm pots containing Levington potting compost, during March 1988, in a bee-proof glasshouse at the Bush Estate , Edinburgh School of Agriculture. Seedlings were singled and staked to prevent lodging. The plants were subjected to minimum day-time and night-time tempertures of 21 and 16°C respectively. Supplementary lighting was provided by 400W sodium lamps suspended 1m above the plants, so achieving a 16 hour photoperiod.

Results

The experiment was arranged as a randomised block, each treatment was replicated 5 times. Treatments are described in Table 6.1.

Table 6.1: Treatments and timings

Treatment	Growth Stage		
	09	31	41
Control	Water + Wetter		
A	EL500		
B	EL500 + BAP		
C	EL500 + BAP	EL500 + BAP	
D	EL500 + BAP		EL500 + BAP
E	EL500 + BAP		BAP + CLIAA (d)
F	EL500 + BAP		BAP + CLIAA (c)

(c) = conventional appliaction (d) = drop leg application

All treatments were applied with a hand-held sprayer, with the drop leg application achieved by spraying the same quantity of chemical upwards from the base of the plant as opposed to the normal application from the top of the plant. All treatments contained the wetter "Agral" at the rate of 1ml / litre.

All flowers were hand-tripped. Pod set, heights, number of harvestable pods and weight of pods and seeds was measured.

Results

Intra-Raceme Percentage Pod Set

Control Plants

Control plants demonstrated the usual pattern of pod set, with the majority of pods set at the proximal three flower positions (average 28.1%), while pod set was reduced at the distal four flower positions (average 4.6%, Fig. 6.1a).

Effect of EL500

Percentage pod set was increased, compared to control plants on the proximal three flowers to 39.5% (Fig.6.1a) and to 13.6% on the four distal flower positions. These increases were not significant.

Effect of EL500 + BAP

Treatment B resulted in a slightly decreased percentage pod set at the three proximal flower positions (26.7%, Fig. 6.1b). However, percentage pod set was increased compared to control plants at the distal four positions to 12.0%.

Figure 6.1a: Effect of EL500 and EL500 + BAP applied at growth stage C₁ on intra-raceme percentage pod set. Actual figures are shown in Appendix 6.1b

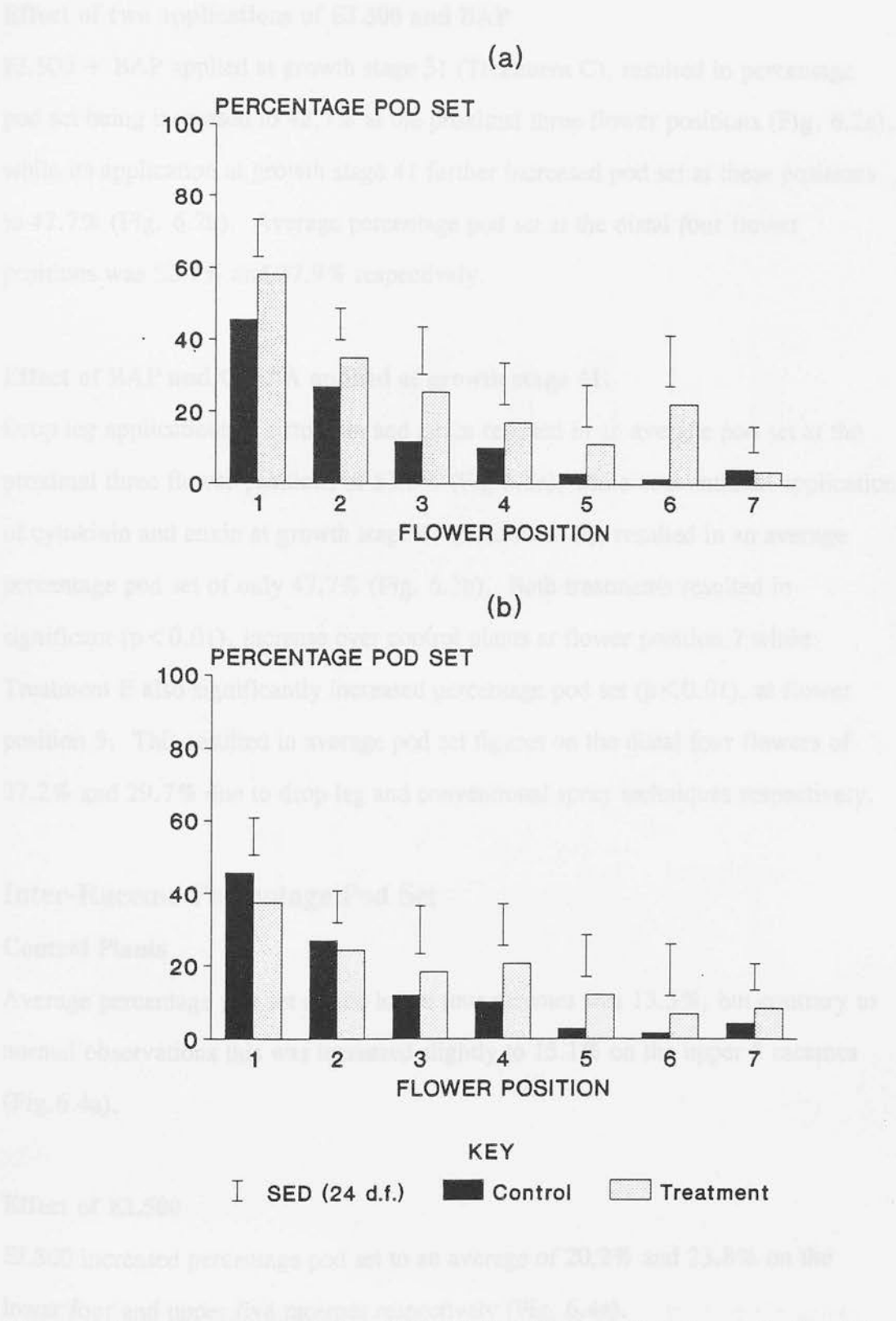


Figure 6.1: Effect of a) EL500 and b) EL500 + BAP applied at growth stage 09, on intra-raceme percentage pod set. (Actual figures are shown in Appendix 6.1)

Effect of two applications of EL500 and BAP

EL500 + BAP applied at growth stage 31 (Treatment C), resulted in percentage pod set being increased to 42.7% at the proximal three flower positions (Fig. 6.2a), while its application at growth stage 41 further increased pod set at these positions to 47.7% (Fig. 6.2b). Average percentage pod set at the distal four flower positions was 32.7% and 37.9% respectively.

Effect of BAP and CLIAA applied at growth stage 41.

Drop leg applications of cytokinin and auxin resulted in an average pod set at the proximal three flower positions of 53.8% (Fig. 6.3a), while conventional application of cytokinin and auxin at growth stage 41 (Treatment F), resulted in an average percentage pod set of only 47.7% (Fig. 6.3b). Both treatments resulted in significant ($p < 0.01$), increase over control plants at flower position 7 while Treatment E also significantly increased percentage pod set ($p < 0.01$), at flower position 5. This resulted in average pod set figures on the distal four flowers of 37.2% and 29.7% due to drop leg and conventional spray techniques respectively.

Inter-Raceme Percentage Pod Set

Control Plants

Average percentage pod set on the lower four racemes was 13.5%, but contrary to normal observations this was increased slightly to 15.1% on the upper 5 racemes (Fig. 6.4a).

Effect of EL500

EL500 increased percentage pod set to an average of 20.2% and 23.8% on the lower four and upper five racemes respectively (Fig. 6.4a).

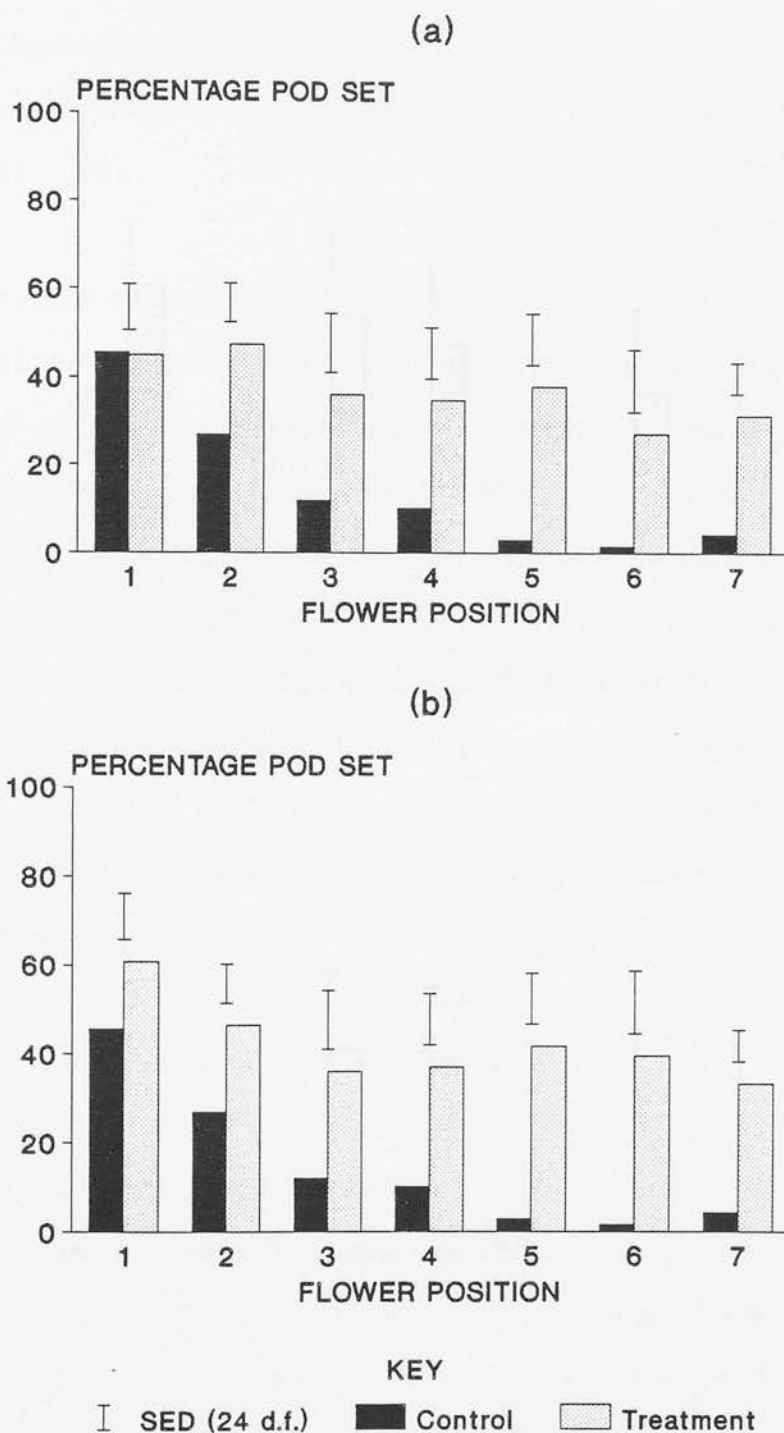


Figure 6.2: Effect of EL500 + BAP applied at a) growth stage 31 and b) growth stage 41 on intra-raceme percentage pod set. (Actual figures are shown in Appendix 6.1)

Effect of ELISA + BAP

Treatment B increased pod set on the lower raceme to 31.3% (Fig. 6.4b).

However, pod set on the upper raceme of the same plant was slightly reduced compared to control plants (Fig. 6.4a).

Effect of two applications of ELISA and BAP

A second application of ELISA and BAP to the same plants 11 days later (average

percentage pod set over the two applications was 31.3% (Fig. 6.5a).

with significant increases in pod set on the lower raceme (Fig. 6.5b) and

2. Application of ELISA and BAP to the same plants 11 days later (average

percentage pod set over the two applications was 31.3% (Fig. 6.5a).

Treatment C significantly increased pod set on the lower raceme compared to control plants at

nodes 5 and 6 (Fig. 6.5b). Treatment D increased pod set on the upper raceme

nodes 5 and 6 (Fig. 6.5a). Treatment E increased pod set over

control plants at nodes 5 and 6 (Fig. 6.5b). Average pod set was increased to

62.3% (Fig. 6.5a).

Effect of BAP + CLIAA

The drop leg spray technique on the lower raceme of the same plants

resulted in 66.6% pod set on the lower raceme (Fig. 6.6a) and 31.3% (Fig. 6.6b).

(Fig. 6.6a). Control plants had 31.3% pod set on the lower raceme compared

to control plants, but to such an extent that average pod set was 31.4% (Fig.

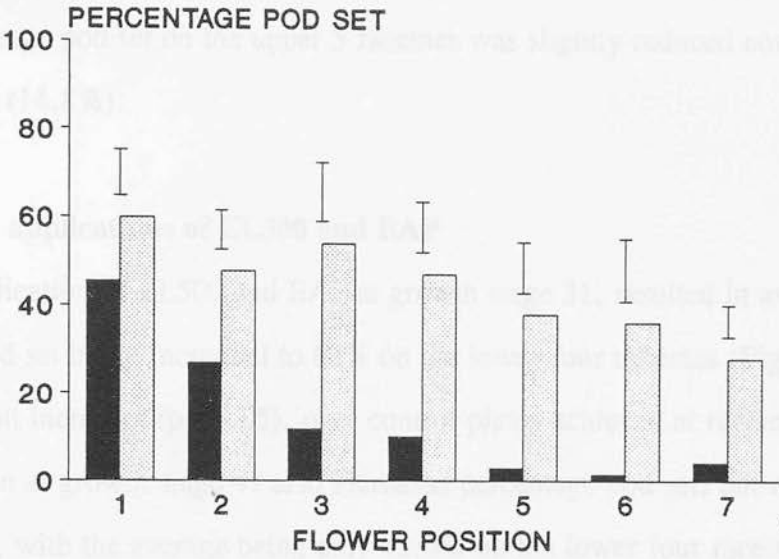
6.6b). Pod set on the upper raceme of the same plants was 31.3% by

Treatment F (Fig. 6.6a). Significant increases in pod set on the lower raceme were

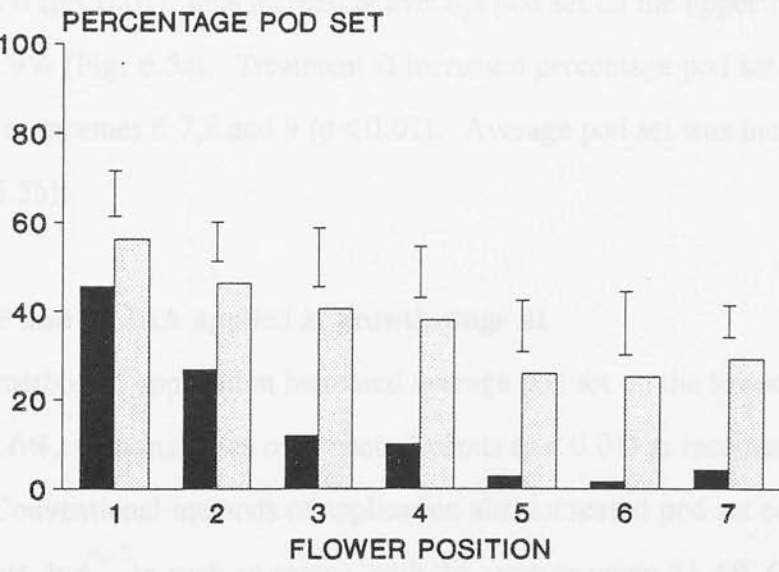
observed at nodes 6, 7 and 8. Treatment F also showed slight increases over

control plants at

(a)



(b)



KEY

┆ SED (24 d.f.) ■ Control ▨ Treatment

Figure 6.3: Effect of BAP and CLIAA applied at growth stage 41, by a) drop leg spray and b) conventional spray techniques, on intra raceme percentage pod set. (Actual figures are shown in Appendix 6.1).

Effect of EL500 + BAP

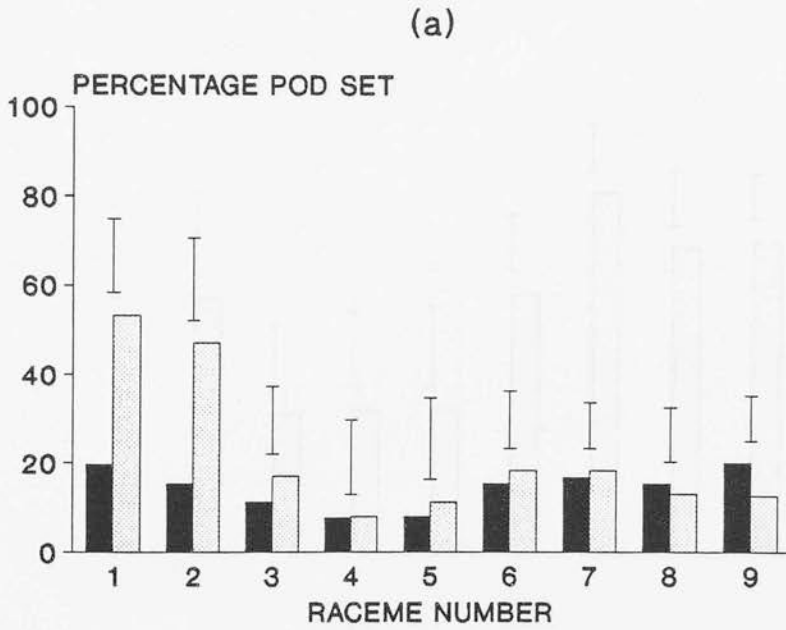
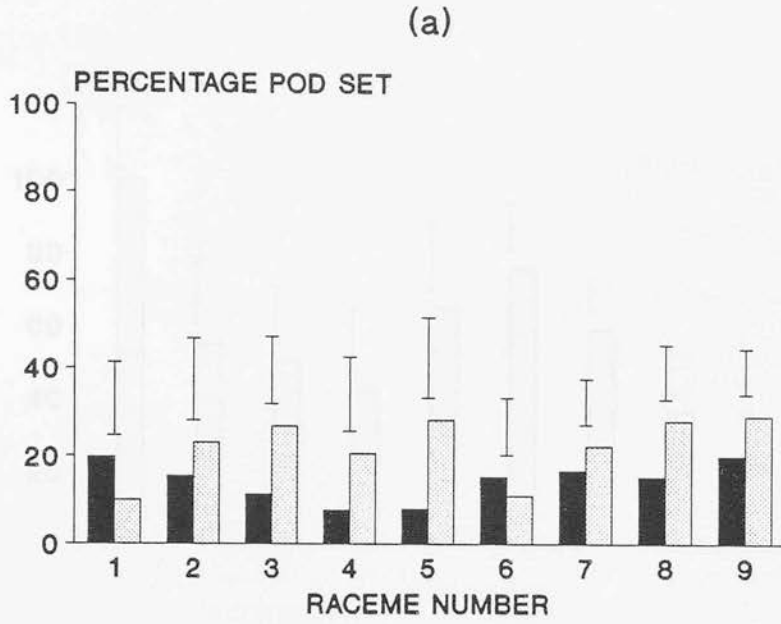
Treatment B increased pod set on the lower four racemes to 31.3% (Fig. 6.4b), however, average pod set on the upper 5 racemes was slightly reduced compared to control plants (14.8%).

Effect of two applications of EL500 and BAP

A second application of EL500 and BAP at growth stage 31, resulted in average percentage pod set being increased to 62% on the lower four racemes (Fig. 6.5a), with significant increases ($p < 0.05$), over control plants achieved at racemes 1 and 2. Application at growth stage 41 also increased percentage pod set, but not to such a great extent, with the average being only 43.6% on the lower four racemes. Treatment C significantly increased percentage pod set compared to control plants at racemes 5 and 6 ($p < 0.01$), thus increasing average pod set on the upper five racemes to 51.9% (Fig. 6.5a). Treatment D increased percentage pod set over control plants at racemes 6, 7, 8 and 9 ($p < 0.01$). Average pod set was increased to 62.5% (Fig. 6.5b).

Effect of BAP and CLIAA applied at growth stage 41

The drop leg method of application increased average pod set on the lower four racemes to 66.6%, with increases over control plants ($p < 0.01$) at racemes 1 and 2 (Fig. 6.6a). Conventional methods of application also increased pod set compared to control plants, but ^{not} to such an extent, with the average being 51.4% (Fig. 6.6b). Pod set on the upper five racemes was increased to an average of 49.6%, by Treatment E (Fig. 6.6a). Significant increases ($p < 0.01$), over control plants were evident at racemes 6, 7 and 8. Treatment F also showed similar increases over control plants, but average percentage pod set was slightly decreased to 48.6%.



KEY

┆ SED (24 d.f.) ■ Control ▨ Treatment

Figure 6.4: Effect of a) EL500 and b) EL500 + BAP applied at growth stage 09, on inter-raceme percentage pod set. (Actual figures are shown in Appendix 6.2)

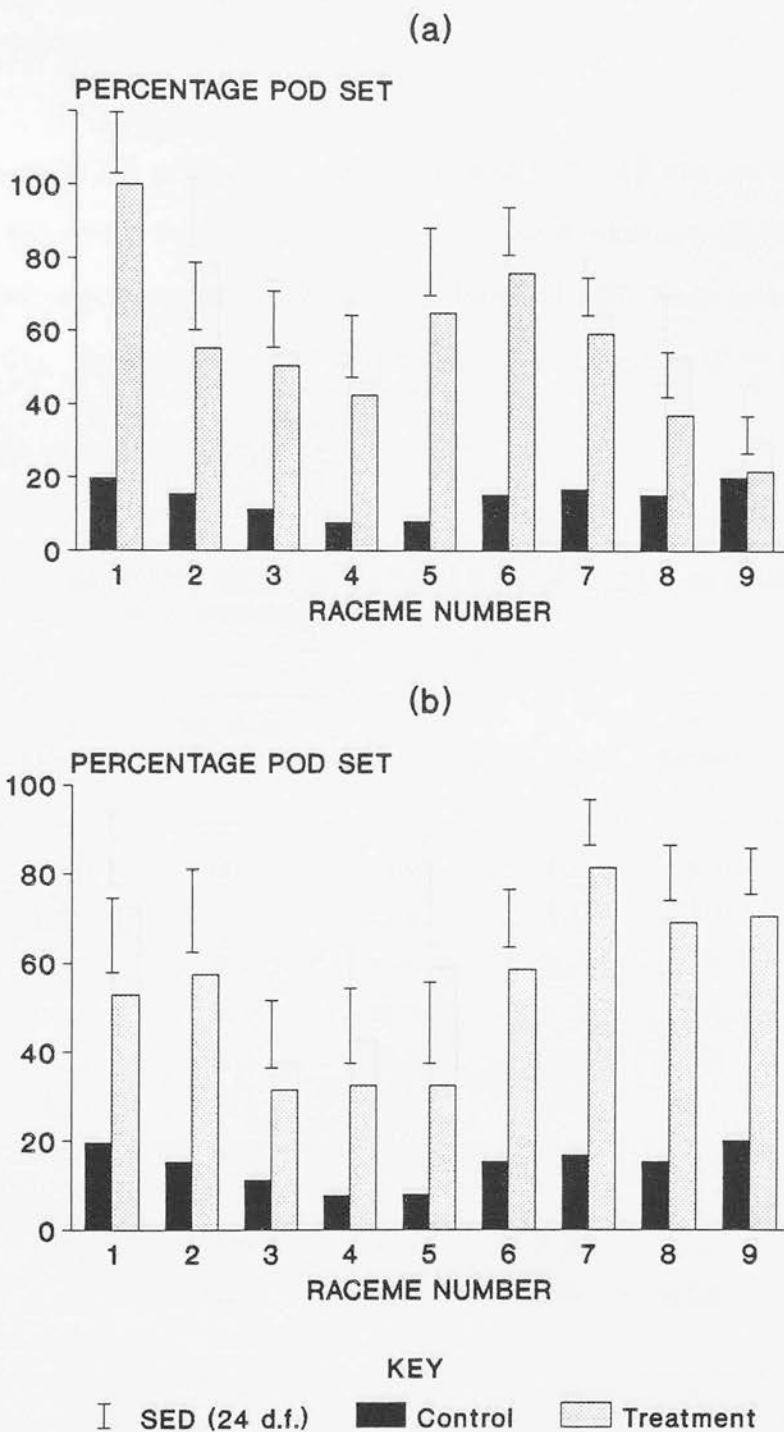
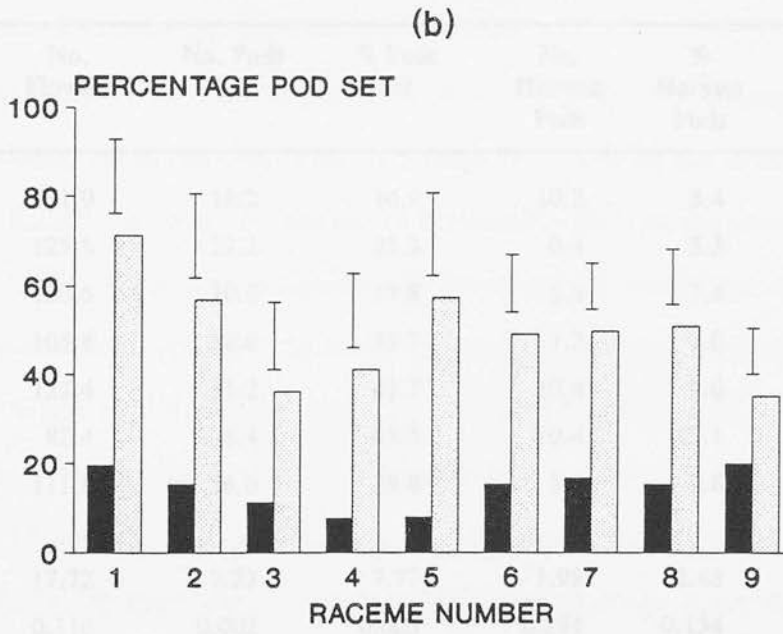
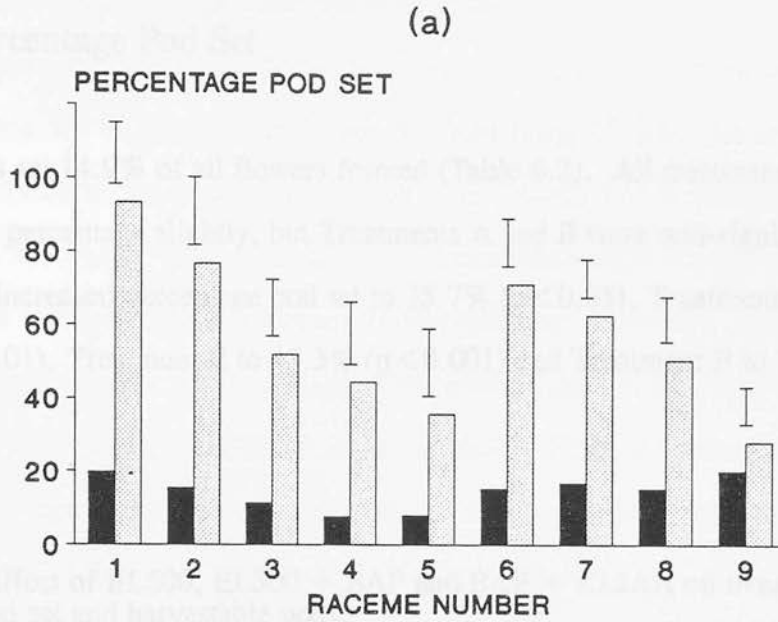


Figure 6.5: Effect of EL500 + BAP applied at a) growth stage 31 and b) growth stage 41 on inter-raceme percentage pod set. (Actual figures are shown in Appendix 6.2)



KEY

SED (24 d.f.) Control Treatment

Figure 6.6: Effect of BAP and CLIAA applied at growth stage 41, by a) drop leg spray and b) conventional spray techniques, on inter-raceme percentage pod set. (Actual figures are shown in Appendix 6.2).

Overall Percentage Pod Set

Control plants set 14.9% of all flowers formed (Table 6.2). All treatments increased this percentage slightly, but Treatments A and B were non-significant. Treatment C increased percentage pod set to 35.7% ($p < 0.05$), Treatment D to 41.7% ($p < 0.01$), Treatment E to 45.3% ($p < 0.001$) and Treatment F to 35.8% ($p < 0.01$).

Table 6.2: Effect of EL500, EL500 + BAP and BAP + CLIAA on overall percentage pod set and harvestable pods.

Treatment	No. Flowers	No. Pods Set	% Pods Set	No. Harvest Pods	% Harvest Pods	% Pod Retention
Control	131.0	18.2	14.9	10.2	8.4	56.9
A	125.8	27.2	23.3	6.4	5.5	26.1
B	130.6	20.8	17.8	8.4	7.4	47.2
C	105.8	38.0	35.7	7.2	7.0	20.5
D	122.4	51.2	41.7	10.4	9.0	23.7
E	82.4	36.4	45.3	10.4	13.1	29.4
F	111.8	36.6	35.8	8.6	7.8	29.1
SED (24 df)	17.72	7.22	7.77	1.98	2.48	10.20
f value	0.116	0.002	0.003	0.284	0.134	0.012

All figures represent the mean of 5 plants

Intra-Raceme Percentage Harvestable Pods

On control plants the majority of harvestable pods were on flower position 1, with the average over the three proximal flower positions being 17.3%. As shown in previous experiments, percentage harvestable pods at distal flower position was reduced to an average of 1.5% (Fig. 6.7a). No treatments significantly increased the percentage of harvestable pods at any flower position. However, all treatments slightly increased the average percentage of harvestable pods at the distal four flower positions to between 1.6-5.2% (Figs. 6.7-6.9). The only treatment to increase percentage harvestable pods at proximal flower positions was Treatment E (Fig. 6.9a), to 22.1%.

Inter-Raceme Percentage Harvestable Pods

As in previous experiments, control plants exhibited the greatest percentage of harvestable pods at lower racemes, with an average of 11.6% on the lower four racemes (Fig. 6.10a). Average percentage of harvestable pods on the upper five racemes was 1.0%. No treatment significantly increased the percentage of harvestable pods on the upper five racemes, however, in all cases it was increased to between, 9.1 and 14.7% (Figs. 6.10-6.12). Treatment E (Fig. 6.12a), increased ($p < 0.01$) the percentage of harvestable pods at raceme 3, causing the average percentage of harvestable pods on the lower four racemes to be increased to 24.3%. Treatment E (drop-leg application), also increased the percentage of harvestable pods ($p < 0.05$) at raceme 1, compared to the conventional method of spray application (Treatment F).

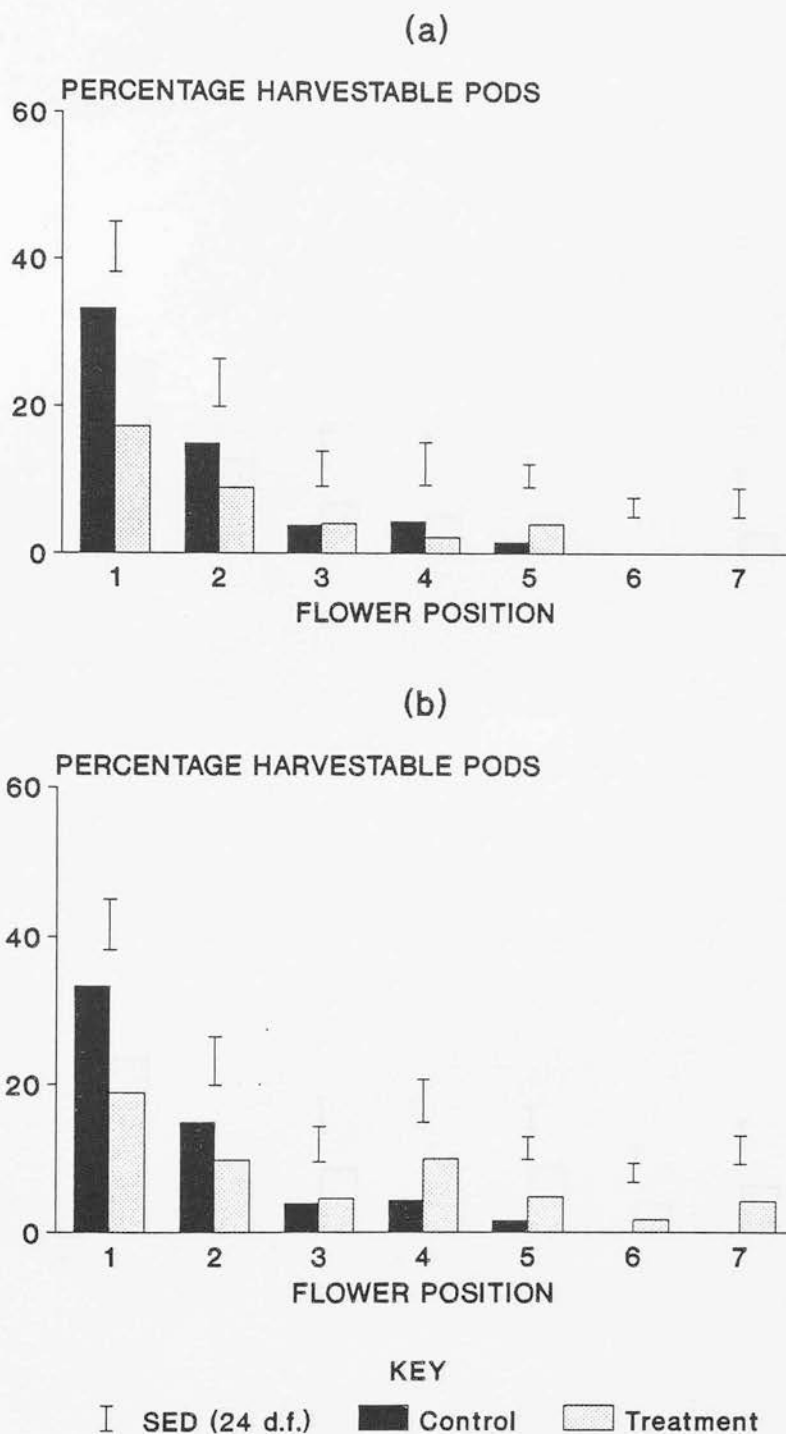


Figure 6.7: Effect of a) EL500 and b) EL500 + BAP applied at growth stage 09, on intra-raceme percentage harvestable pods. (Actual figures are shown in Appendix 6.3)

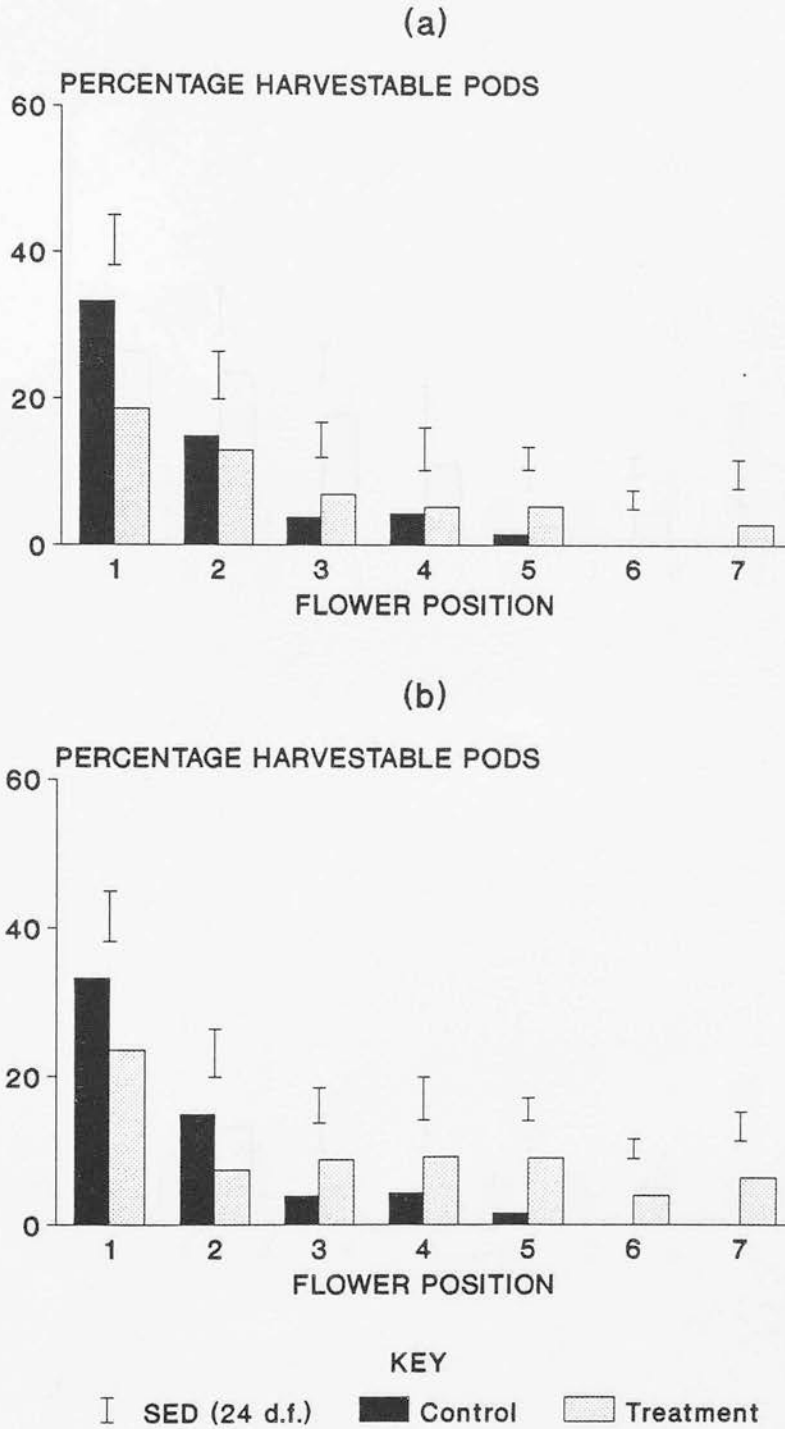


Figure 6.8: Effect of EL500 + BAP applied at a) growth stage 31 and b) growth stage 41 on intra-raceme percentage harvestable pods. (Actual figures are shown in Appendix 6.3)

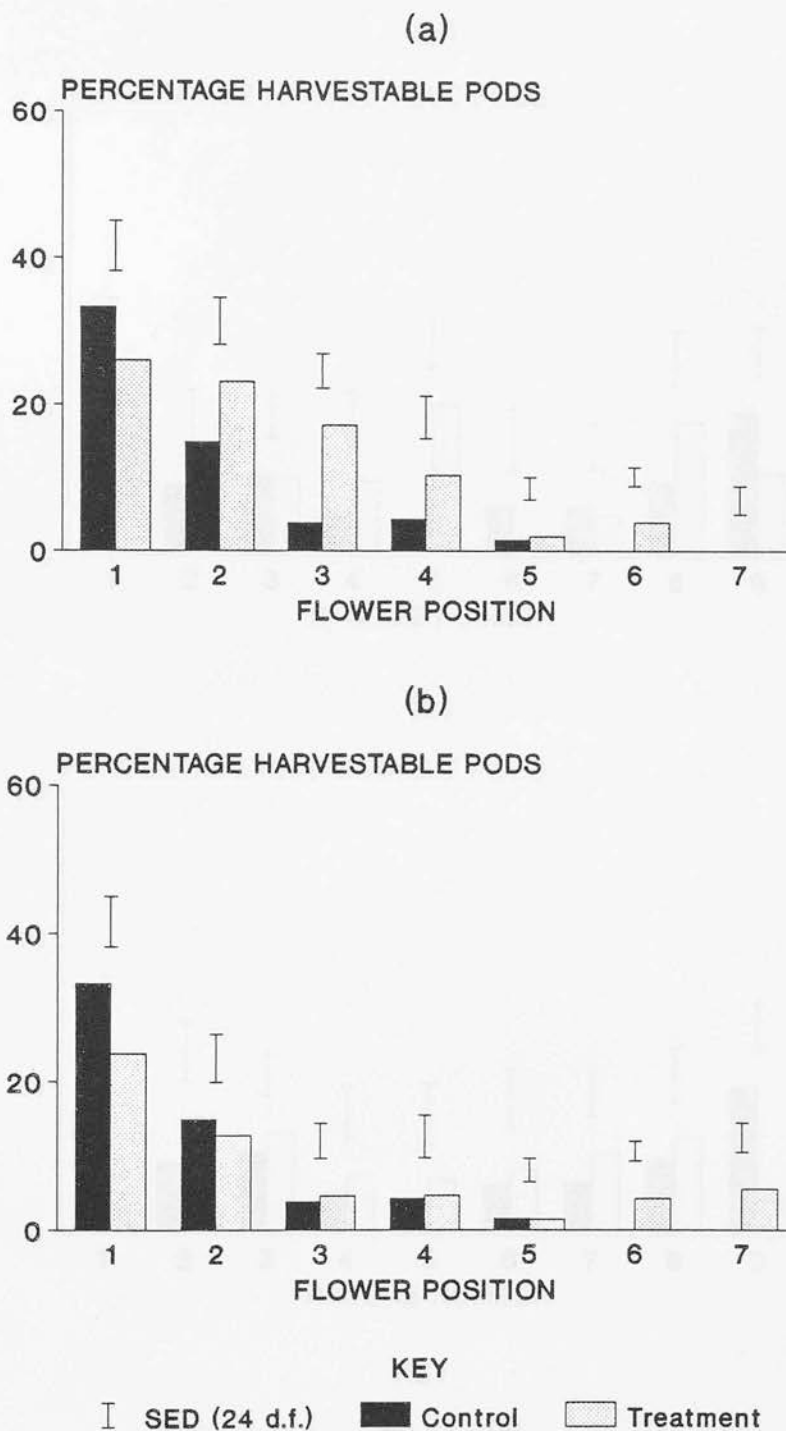


Figure 6.9: Effect of BAP + CLIAA applied at growth stage 41, by a) drop leg spray and b) conventional spray techniques, on intra-raceme percentage harvestable pods. (Actual figures are shown in Appendix 6.3).

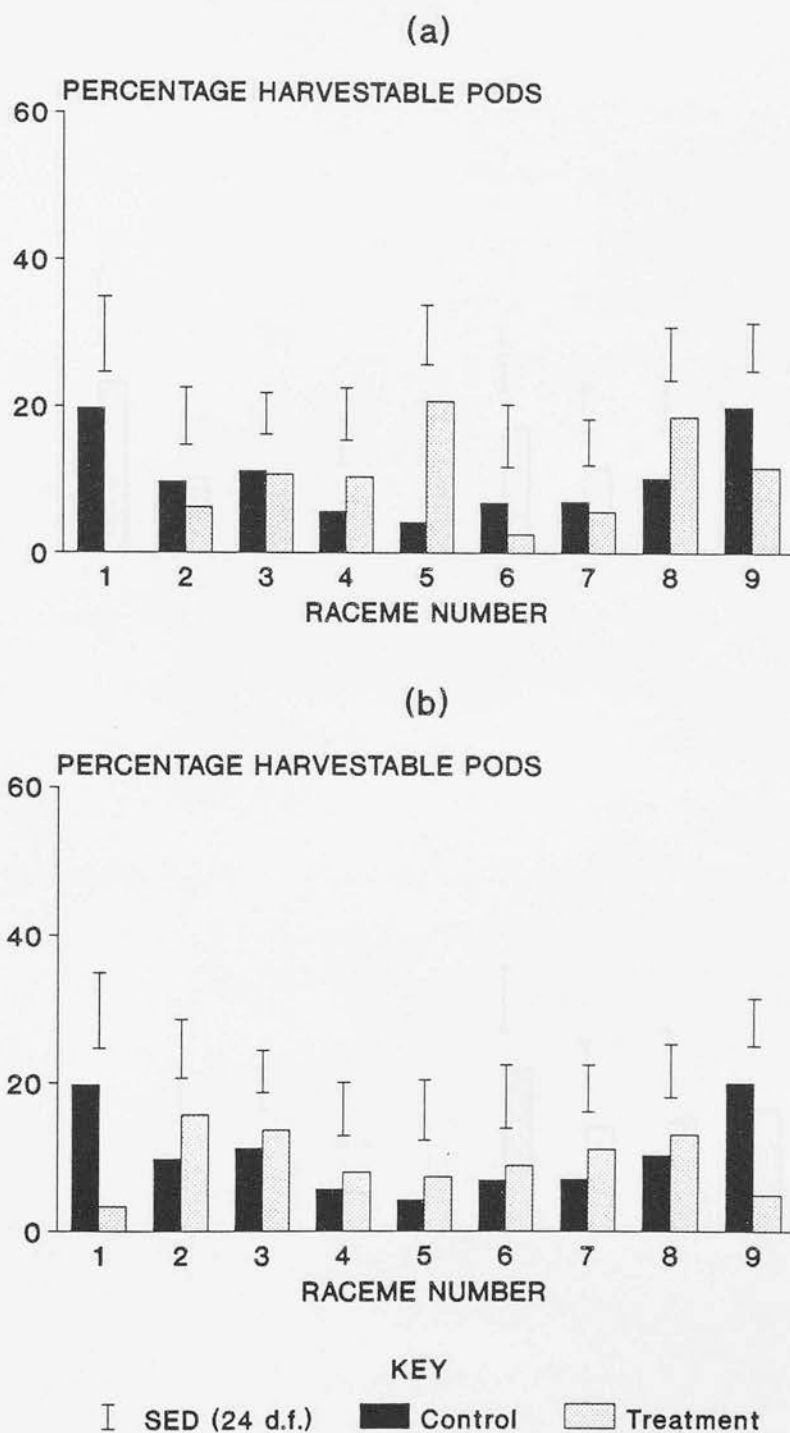


Figure 6.10: Effect of a) EL500 and b) EL500 + BAP applied at growth stage 09, on inter-raceme percentage harvestable pods. (Actual figures are shown in Appendix 6.4)

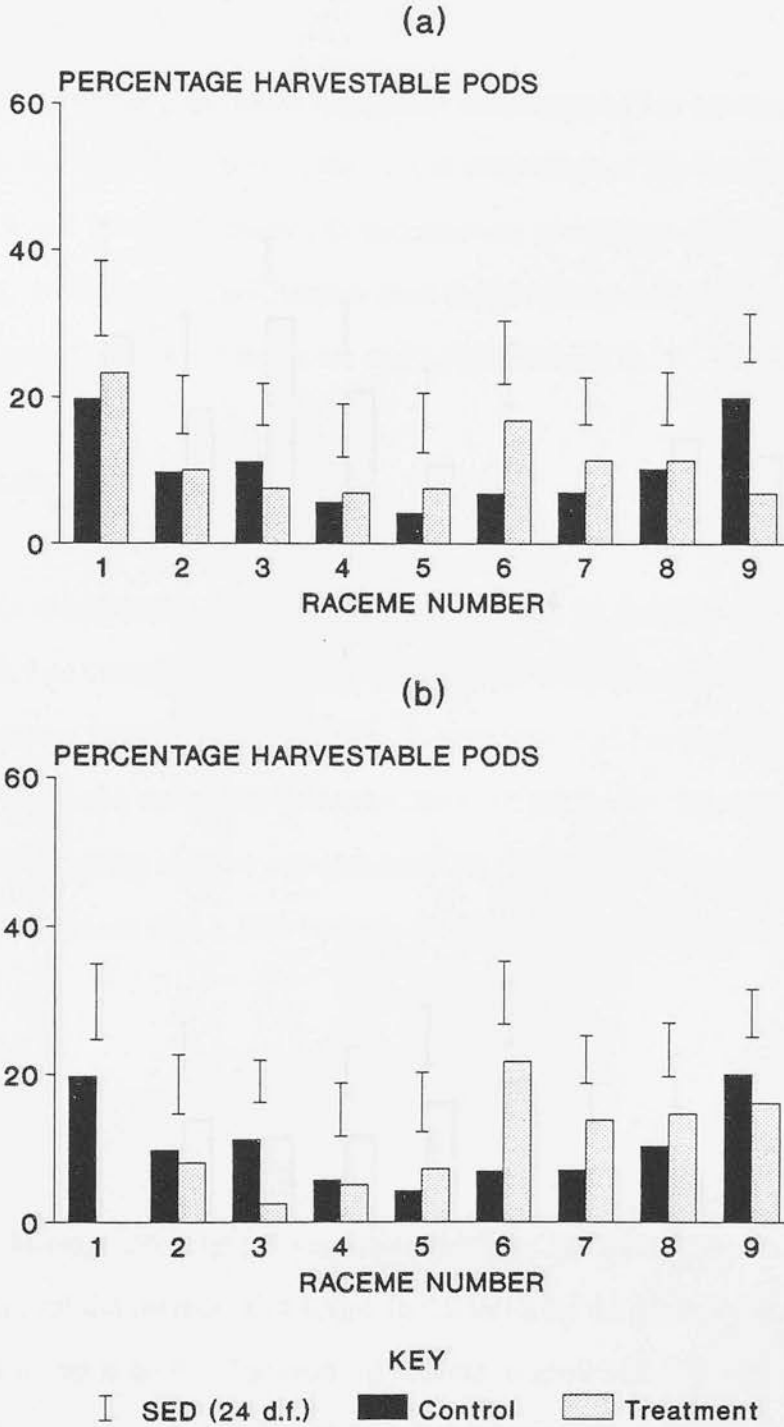


Figure 6.11: Effect of EL500 + BAP applied at a) growth stage 31 and b) growth stage 41, on inter-raceme percentage harvestable pods. (Actual figures are shown in Appendix 6.4)

Overall Percentage Harvestable Pods

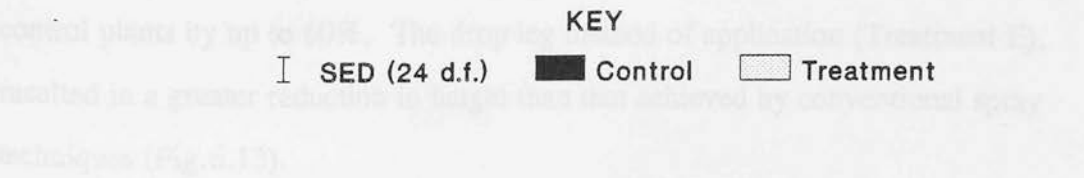
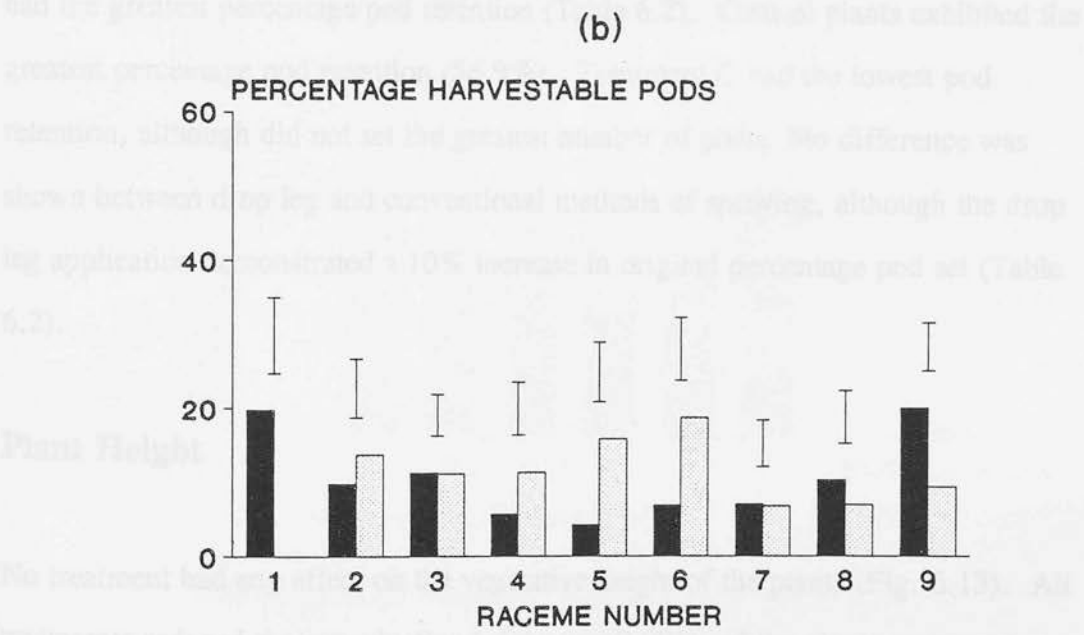
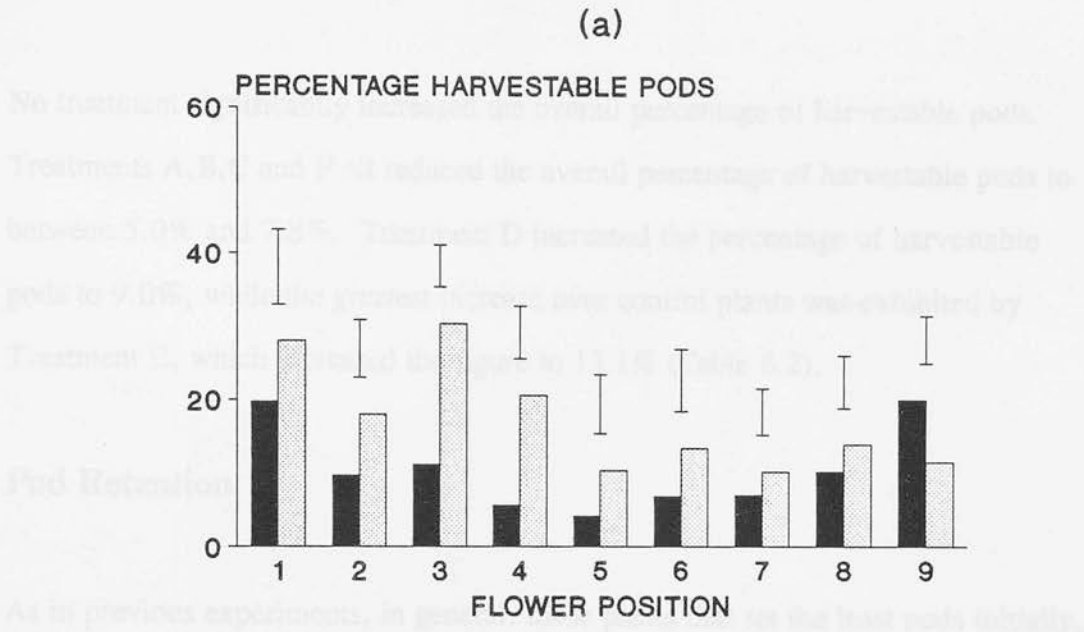


Figure 6.12: Effect of BAP + CLIAA applied at growth stage 41, by a) drop leg spray and b) conventional spray techniques, on inter-raceme percentage harvestable pods. (Actual figures are shown in Appendix 6.4)

Overall Percentage Harvestable Pods

No treatment significantly increased the overall percentage of harvestable pods. Treatments A,B,C and F all reduced the overall percentage of harvestable pods to between 5.0% and 7.8%. Treatment D increased the percentage of harvestable pods to 9.0%, while the greatest increase over control plants was exhibited by Treatment E, which increased the figure to 13.1% (Table 6.2).

Pod Retention

As in previous experiments, in general, those plants that set the least pods initially, had the greatest percentage pod retention (Table 6.2). Control plants exhibited the greatest percentage pod retention (56.9%). Treatment C had the lowest pod retention, although did not set the greatest number of pods. No difference was shown between drop leg and conventional methods of spraying, although the drop leg application demonstrated a 10% increase in original percentage pod set (Table 6.2).

Plant Height

No treatment had any effect on the vegetative height of the plants (Fig. 6.13). All treatments reduced the reproductive height ($p < 0.001$), of the plants compared to control plants by up to 60%. The drop leg method of application (Treatment E), resulted in a greater reduction in height than that achieved by conventional spray techniques (Fig.6.13).

Figure 6.13: Effect of EL900, DAP and CLIAA, applied by drop leg and conventional spray techniques on vegetative and reproductive plant height. (Actual figures are shown in Appendix 6.2)

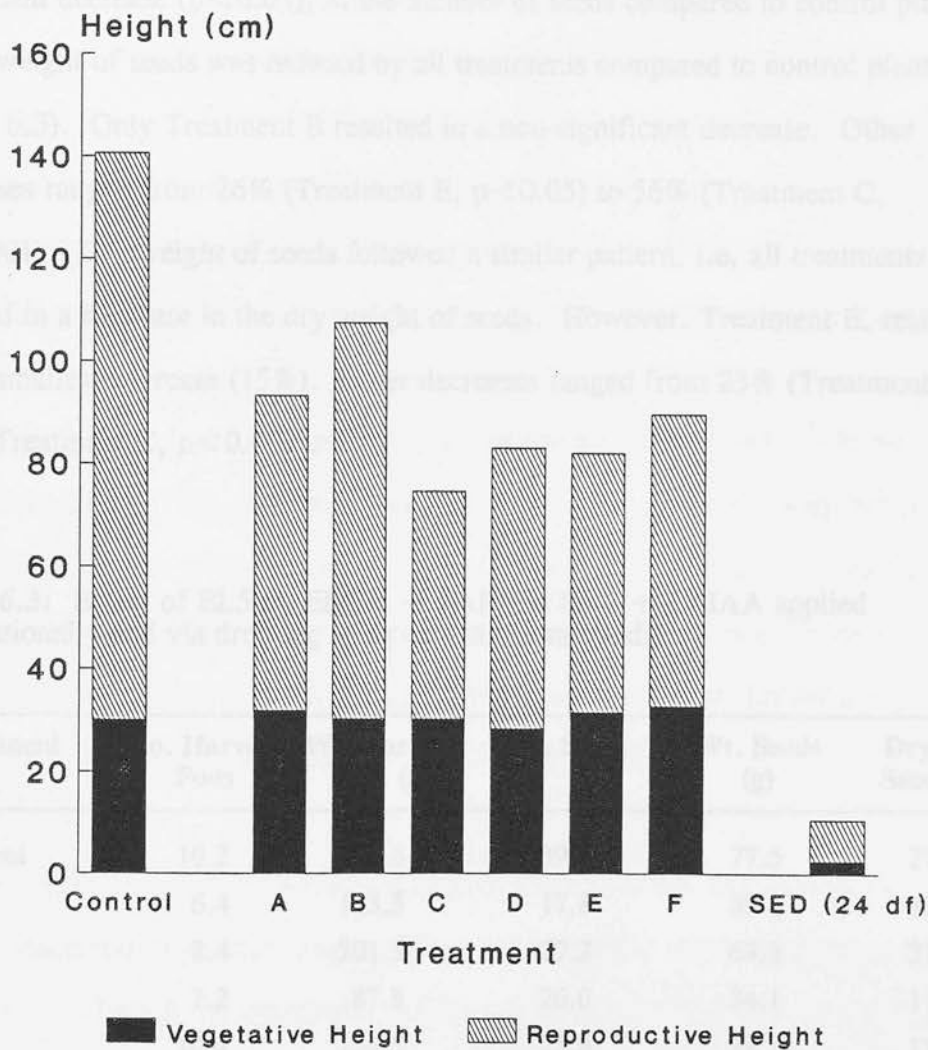


Figure 6.13: Effect of EL500, BAP and CLIAA, applied by drop leg and conventional spray techniques on vegetative and reproductive plant height. (Actual figures are shown in Appendix 6.5)

Mainstem Yield

No treatment had any effect on the number of harvestable pods contained on the mainstem. However, all treatments except Treatment B, resulted in a decrease ($p < 0.01$) in fresh weight of pods. All treatments apart from B and E, resulted in a significant decrease ($p < 0.05$), in the number of seeds compared to control plants. Fresh weight of seeds was reduced by all treatments compared to control plants (Table 6.3). Only Treatment B resulted in a non-significant decrease. Other decreases ranged from 26% (Treatment E, $p < 0.05$) to 56% (Treatment C, $p < 0.001$). Dry weight of seeds followed a similar pattern, i.e. all treatments resulted in a decrease in the dry weight of seeds. However, Treatment E, resulted in the smallest decrease (15%). Other decreases ranged from 23% (Treatment B) to 59% (Treatment C, $p < 0.001$).

Table 6.3: Effect of EL500, EL500 + BAP and BAP + CLIAA applied conventionally and via drop leg spray on mainstem yield.

Treatment	No. Harvest Pods	Wt. Harvest Pods (g)	No. Seeds	Wt. Seeds (g)	Dry Wt. Seeds (g)
Control	10.2	229.6	39.2	77.5	27.4
A	6.4	113.3	17.8	39.1	12.0
B	8.4	201.3	27.2	63.6	21.3
C	7.2	87.8	20.0	34.1	11.4
D	10.4	126.5	25.0	39.3	11.7
E	10.4	127.0	28.0	57.7	23.4
F	8.6	149.0	26.0	53.5	19.4
SED (24 df)	1.98	26.58	5.86	9.12	3.89
f value	0.284	<0.001	0.035	<0.001	0.001

All figures represent the mean of 5 plants

Discussion

As in previous experiments, all treatments increased pod set compared to control plants. The application of EL500 at growth stage 09 resulted in an overall pod set of 23.3%, which directly corresponds to the 23% reported in Chapter 5.

Application of BAP at growth stage 09 with EL500 reduced pod set to 17%. A second application of BAP with EL500 increased pod set and this effect was shown to be greater when applied at growth stage 41 rather than at growth stage 31. BAP applied at growth stage 41 in conjunction with CLIAA resulted in the greatest amount of pod set, especially if the chemical was applied via a drop leg sprayer.

Control plants had a greater number of harvestable pods than those in Chapter 5 (10.2 compared to 7.4). This may therefore, be part of the reason why fewer significant increases were evident at harvest. However, the greatest number of pods per plant were on plants subjected to applications of EL500 + BAP or BAP + CLIAA at growth stage 41 (drop leg). Of these two treatments, the drop leg method of application had a greater amount of pod retention.

It was suggested in Chapter 4, that the application of CLIAA and GA_3 to the flowers increased the overall height of the plant especially when applied pre-pollination. Thus, if the chemicals could be applied to the area where the pods had already set (i.e. the area where the PGSs could be directly utilised by the pods), rather than to the apex of the plant, then this phenomena would be slightly reduced. It was shown in this experiment that application of CLIAA via the drop leg method resulted in a shorter plant than those treated by the traditional method of spraying (Fig. 6.13). It was also shown in Chapter 4, that the application of CLIAA after fertilization was important for vascular differentiation. It would appear from the results gained in this experiment, that when the chemical is applied at growth stage 41, that it aids pod retention especially on racemes 3 and 4 (Figs. 6.3 and 6.6), so

resulting in a greater number of harvestable pods. However, this effect is greater when the chemical is applied to the area where the pods are forming (by drop leg) and not when applied to the apex. This suggests that CLIAA can be "locked up" in apex development when applied conventionally, which in turn can lead to taller and more indeterminate plants with fewer harvestable pods.

It was found from this experiment, that the application of more plant growth substances at a later timing can aid pod retention. This may be because the application of cytokinins stimulate cell division (Moore, 1979) and auxins stimulate cell expansion (Moore, 1979). Consequently, the sink size is increased and so assimilates are more actively attracted (Wareing, 1978). However, it would also appear from this experiment that either the effects due to the applied plant growth substances (BAP + CLIAA), are transient, or that individual plants tend to support only a certain number of pods, i.e. assimilates eventually become limiting so resulting in late pod abortion, small pods and dominance at the traditional proximal flower positions. It would be logical therefore, to try and increase assimilate production. This may be possible by either stimulating nodule formation or by adding artificial fertilizer. In Chapter 5 it was demonstrated that the addition of anti-gibberellins reduced the amount of root nodules. In addition, as the amount of dry matter increased in root nodules, the amount of dry matter in the beans decreased. Thus, it would be logical to add artificial fertilizer to the growing plant in order to try and boost assimilate production during pod fill.

Introduction

It was found in Chapter 6 that the growth of *Phaseolus vulgaris* L. is growth stage 4, added good retention. However, the growth stage 4 is not the best stage for the growth of the plant. This may be due to the fact that the growth stage 4 is not the best stage for the growth of the plant.

Chapter 7

Effect of EL500, BAP, CLIAA, fertilizer and density

production, root quantity and overall yield

Using (1973a), found that when plants were grown at low density (36 plants/m²), the number of roots per plant increased by 40% when the density was increased to 45 plants/m², while root yield per plant decreased by 40%. However, yield per unit area was increased by 30% at low density. Smith (1973) observed that as plant population increased, root quantity per plant decreased as well as root yield per unit area. However, root yield per unit area was increased by 30% at low density. This is due to the fact that the number of roots per plant decreased as the density increased, but the number of plants per unit area increased. This is due to the fact that the number of roots per plant decreased as the density increased, but the number of plants per unit area increased.

The aim of this experiment was to determine the effect of plant growth regulators (EL500, BAP, CLIAA) and fertilizer on the growth and yield of *Phaseolus vulgaris* L. The results of the experiment showed that the application of EL500, BAP, and CLIAA significantly increased the growth and yield of the plant. The application of fertilizer also significantly increased the growth and yield of the plant. The results of the experiment showed that the application of EL500, BAP, and CLIAA significantly increased the growth and yield of the plant. The application of fertilizer also significantly increased the growth and yield of the plant.

Introduction

It was found in Chapter 6, that the application of BAP and CLIAA, at growth stage 41, aided pod retention. However, the final yield of beans was not increased. This may have been due to an inability of the plant to produce enough assimilates to support these extra pods. Hebblethwaite (1970), suggested that only 60% of U.K. bean crops received any form of artificial fertilizer. It was suggested in previous chapters that a small addition of nitrogen at early pod set, may aid assimilate production, pod retention and overall yield

Ishag (1973a), found that when plants were grown at low densities (36 plants m^{-2}), the number of pods per plant increased by 40% compared to plants grown at a high density (57 plants m^{-2}), while seed yield per plant increased by 50%. However, yield per unit area was increased by 33% in dense crops. Smith (1982), discovered that as plant population increased, plants became more pre-disposed to set pods at higher racemes, although pods still remained at proximal flower positions. Flower abscission was increased at higher densities due shading and resulting reduction in assimilate production.

The aim of this experiment was to determine a) ideal plant populations for the crop when treated with plant growth substances and to assess whether artificial fertilizer could increase overall yield. In addition, the layout of the experiment allowed comparisons to the previous year's field trial, thus giving some continuity of results.

Method

Seeds of Threefold White were sown on at the Bush Estate, Edinburgh School of Agriculture on 11 May 1988. The experiment was designed as a randomised block layout, each treatment being replicated 3 times. Each plot measured 1 x 1.5m and seeds were sown at regular spacings along 30cm rows in order to achieve plant populations of 10, 15, 20, 25, 30 and 40 plants m⁻². A distance of 1m was left between plots.

The previous crop was barley. Soil samples indicated that the indices for P and K were 2. No compound fertilizer was placed in the seed bed. No herbicides were applied to the growing crop. However, the stale seed bed was sprayed with Gramoxone (paraquat), prior to cultivations. Plots were kept weed free during the growing period by hoeing. Chocolate spot was treated at growth stage 31, with Benlate (benomyl), at the rate of 1kg/ha.

Growth substances used were: EL500 (flurprimidol), at the rate of 0.25g/litre, BAP (6-benzylaminopurine), at the rate of 23mg/litre and CLIAA (4-chloroindole), at the rate of 50mg/litre. On the applicable plots (Table 7.1), compound fertilizer (20.10.10), was added at the rate of 50.25.25 kg/ha. All applications of chemical at growth stage 09 were made with a Cooper Pegler knapsack sprayer fitted with fan-jet nozzles and were applied to run off. This represented an application of 1500 litres/ha. At growth stage 41, the applications were again with a Cooper Pegler knapsack, but fitted with an extension lance with a double-swivel hollow cone jet, directed upwards, so imitating a drop-leg technique of application. Application rate was 1500 litres/ha. All treatments contained the non-ionic wetter "Agral", at the rate of 1ml/litre. Control plants were sprayed with a mixture of water and wetter at growth stage 09.

Table 7.1: Treatments

Treatment	Growth Stage		
	09	31	41
Control	Water + Wetter		
A	EL500		
B	EL500 + BAP		BAP + CLIAA
C	EL500 + BAP	Fertilizer	BAP + CLIAA

In order to prevent edge effects, 5 plants were chosen at random from the centre of each plot and tagged to allow accurate identification. Mainstems of these plants were scored for number of flowers formed and pod set. At harvest (19-20 September), the same plants were scored for height, number of tillers, number of harvestable pods, weight of harvestable pods and weight of seeds on a mainstem, tiller and whole plant basis. Plants were then further dissected into component parts of stem and leaf, pods and beans. These parts were then weighed and placed into individual polythene bags and frozen, until dry weights could be carried out. Dry weights were achieved by placing the sample into an oven set at 90°C until the weights reached equilibrium.

Results

Intra-Raceme Percentage Pod Set

Effect Of PGS

As in previous experiments, control plants set the majority of pods at proximal flower positions on the raceme. The average for the first three flower positions

being 29.9%, while average pod set on the distal three flowers averaged 1.7% (Table 7.2).

The application of EL500 at growth stage 09 (Treatment A), resulted in significant ($p < 0.05$), increases in percentage pod set at flower positions 1, 2 and 3, resulting in the average pod set for the proximal three flowers being 40.6%. Increases ($p < 0.001$), were also evident at flower 4 over control plants, which meant the average for the distal three flowers was 5.5% (Table 7.2).

The application of EL500 + BAP at growth stage 09, followed by BAP + CLIAA at growth Stage 41 (Treatment B), resulted in increases ($p < 0.05$) compared to control plants at all flower positions (Table 7.2). Consequently, average percentage pod set at the proximal flower positions was 50.8%, while at the distal three flower positions it was 9.8%. In addition, increases were also evident over EL500 at flower positions 1 and 5 ($p < 0.05$) and 2, 3 and 4 ($p < 0.001$).

The addition of fertilizer at growth stage 31 (Treatment C), resulted in similar percentage pod set figures to those receiving no fertilizer (Treatment B), i.e. on the proximal three flowers it was 47.5% and 10.3% on the distal three flowers (Table 7.2).

Effect of Density

Density had no effect on the percentage of pods set at any flower position apart from flower position 5. At this position, percentage pod set at 25 plants m^{-2} was significantly lower ($p < 0.05$) than in plant populations of 10, 20 and 40 plants m^{-2} . No difference was seen between density and plant growth regulator treatment (Table 7.2).

Table 7.2: Effect of EL500, BAP + CLIAA, Fertilizer and Density on intra-raceme percentage pod set.

Treatment	Density (m ⁻²)	Flower Position					
		1	2	3	4	5	6
Control	10	52.2	27.9	9.9	4.6	0.7	0.0
	15	58.2	33.0	10.9	5.8	1.8	0.0
	20	51.4	35.1	8.9	4.5	0.0	0.0
	25	51.3	28.2	5.9	2.2	0.0	0.0
	30	49.8	34.9	9.8	6.1	0.0	0.0
	40	44.1	20.2	6.9	3.8	0.0	1.1
<i>Mean</i>		<i>51.2</i>	<i>29.9</i>	<i>8.7</i>	<i>4.5</i>	<i>0.4</i>	<i>0.2</i>
A	10	57.2	47.8	23.3	14.5	6.1	2.6
	15	50.0	43.3	26.3	16.4	0.4	0.0
	20	58.1	41.2	25.1	7.7	1.2	0.0
	25	57.9	43.8	22.1	10.9	0.0	1.0
	30	59.0	40.3	14.5	17.4	0.0	0.0
	40	60.2	41.5	18.5	17.7	3.0	0.0
<i>Mean</i>		<i>57.1</i>	<i>43.0</i>	<i>21.6</i>	<i>14.1</i>	<i>1.8</i>	<i>0.6</i>
B	10	58.5	56.0	35.9	20.5	6.1	0.8
	15	63.2	52.4	33.1	18.5	5.4	1.9
	20	60.7	49.3	29.5	27.3	8.7	6.5
	25	60.5	52.2	36.3	19.4	2.7	2.3
	30	65.9	52.2	36.5	15.2	2.4	0.0
	40	74.1	61.9	36.2	21.2	11.6	5.0
<i>Mean</i>		<i>63.8</i>	<i>54.0</i>	<i>34.6</i>	<i>20.4</i>	<i>6.2</i>	<i>2.8</i>
C	10	57.7	55.4	26.9	16.8	8.2	8.6
	15	58.6	53.3	36.3	22.9	5.4	4.6
	20	58.7	43.2	29.1	22.1	9.8	6.6
	25	63.9	42.9	16.4	8.1	0.8	1.1
	30	63.8	51.1	32.9	20.0	8.2	2.4
	40	70.4	61.5	34.4	23.2	8.4	7.5
<i>Mean</i>		<i>62.2</i>	<i>51.2</i>	<i>29.3</i>	<i>18.9</i>	<i>6.8</i>	<i>5.2</i>
Density Mean	10	56.4	46.8	24.0	14.1	5.3	3.0
	15	57.5	45.5	26.7	15.9	3.3	1.6
	20	57.2	42.2	23.1	15.4	4.9	3.3
	25	58.4	41.7	20.2	10.2	0.9	1.1
	30	59.6	44.6	23.4	14.7	2.7	0.6
	40	62.2	46.3	24.0	16.5	5.7	3.4
SED							
PGS		2.64	2.77	2.60	2.27	1.28	1.24
Density		3.24	3.40	3.19	2.77	1.57	1.52
PGS x Density		6.48	6.81	6.38	5.54	3.15	3.04
f value							
PGS (90 df)		<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Density (60 df)		0.521	0.572	0.519	0.251	0.017	0.273
PGS x Density (15 df)		0.324	0.167	0.375	0.434	0.413	0.724

Inter-Raceme Percentage Pod Set

Effect of PGS

Control plants followed the usual pattern of pod set, with the greatest pod set at lowers racemes (Table 7.3). Average percentage pod set on the lower four racemes was 28.5% and on the upper four racemes was 13.3%.

Treatment A, resulted in increased pod set ($p < 0.01$), at the four lower racemes, leading to average pod set being 42.7%. Increases were also evident at the upper four racemes compared to control plants ($p < 0.05$), with average percentage pod set being 22.4% (Table 7.3).

Treatment B, resulted in significant increases ($p < 0.001$), at all racemes over control plants. Average percentage pod set on the lower four racemes was 55.3% and on the upper four racemes was 26.8% (Table 7.3). Increases were also evident over a single EL500 application ($p < 0.01$) on the four lower racemes.

The addition of fertilizer at growth stage 31 (Treatment C), resulted in percentage pod set figures that were slightly less than Treatment B, at all racemes, although, this was not significant. Percentage pod set was increased at all racemes ($p < 0.001$) compared to control plants. Average pod set on the lower four racemes was 52.2% and 23.1% on the upper four racemes (Table 7.3).

Effect of Density

Density only affected percentage pod set on raceme 1. Plant populations of 25 plants m^{-2} had a low percentage pod set, which was significantly lower ($p < 0.05$), than those populations of 10, 15, 30 and 40 plants m^{-2} (Table 7.3).

Table 7.3: Effect of EL500, BAP, CLIAA, Fertilizer and Density on inter-raceme percentage pod set.

Treatment	Density (m ⁻²)	Raceme Number							
		1	2	3	4	5	6	7	8
Control	10	39.5	33.9	21.4	16.6	12.6	13.2	11.0	12.7
	15	43.8	36.9	24.2	18.3	19.8	19.9	15.8	16.6
	20	40.4	34.2	33.4	22.7	24.8	14.8	13.0	8.2
	25	26.9	35.0	25.4	15.6	16.7	13.5	10.1	4.3
	30	41.6	31.7	25.3	23.8	18.0	12.9	14.3	11.7
	40	33.7	24.5	19.1	17.1	10.6	10.3	6.6	7.3
Mean		37.6	32.7	24.8	19.0	17.1	14.1	11.8	10.1
A	10	50.6	46.0	46.4	30.1	33.1	24.4	26.5	18.2
	15	62.8	51.6	38.6	34.8	31.8	23.0	23.1	15.5
	20	48.2	46.3	40.2	34.0	23.7	16.6	16.6	10.9
	25	50.1	47.4	34.9	31.1	28.2	25.0	23.3	14.0
	30	55.3	50.9	35.7	27.3	28.6	22.9	10.9	15.9
	40	55.8	40.0	35.4	30.3	30.6	26.4	16.2	33.3
Mean		53.8	47.0	38.5	31.3	29.3	23.0	19.4	18.0
B	10	80.0	70.2	35.5	43.6	35.3	31.0	26.3	23.3
	15	74.0	51.2	40.8	39.1	35.5	26.5	25.7	25.3
	20	64.4	61.5	57.8	42.1	35.3	28.0	20.3	25.3
	25	69.5	69.2	44.3	38.7	25.2	20.2	16.0	14.5
	30	74.1	62.5	48.5	32.8	26.5	26.6	29.8	18.6
	40	76.4	63.7	53.5	33.5	35.9	30.0	34.2	26.8
Mean		73.1	63.1	46.7	38.3	32.3	27.0	25.4	22.3
C	10	66.8	66.6	43.4	34.0	29.6	29.2	20.6	19.3
	15	80.7	62.7	52.4	43.0	31.9	26.8	18.3	22.9
	20	64.4	63.3	45.8	40.0	31.5	17.9	11.0	19.6
	25	54.1	49.9	36.0	24.4	24.1	17.6	20.4	12.6
	30	63.0	58.6	47.2	38.9	33.0	20.9	14.7	21.9
	40	70.6	63.8	45.6	38.0	27.1	33.6	29.2	22.2
Mean		66.6	60.8	45.1	36.4	29.5	24.3	19.0	19.7
Density Mean	10	59.2	54.2	36.7	31.1	27.6	24.4	21.1	18.4
	15	65.3	50.6	39.0	33.8	29.8	24.0	20.7	20.1
	20	54.4	51.3	44.3	34.7	28.9	19.3	15.2	16.0
	25	50.1	50.4	35.2	27.5	23.5	19.1	17.5	11.4
	30	58.5	50.9	39.2	30.7	26.5	20.8	17.4	17.0
	40	59.1	48.0	38.4	29.7	26.0	25.0	21.5	22.4
SED									
PGS		3.20	2.98	2.45	2.19	2.70	2.68	3.06	3.31
Density		3.92	3.65	3.00	2.69	3.31	3.29	3.74	4.05
PGS x Density		7.85	7.31	6.01	5.37	6.61	6.57	7.49	8.10
f value									
PGS (90df)		<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	0.002
Density (60df)		0.005	0.713	0.060	0.081	0.480	0.242	0.455	0.125
PGS x Density (15df)		0.723	0.216	0.026	0.247	0.648	0.710	0.282	0.854

Only at raceme 3, was there any interaction between PGS and density. As the density and number of PGS applications increased, the trend was for percentage pod set to increase.

Overall Pod Set

Effect of PGS

Contrary to previous results, the application of plant growth substances had a significant effect on the number of flowers per mainstem. In control plants, the average number was 62.8 (Table 7.4), this was reduced ($p < 0.05$), to 55.5 by Treatment C. The average number of pods per mainstem was increased significantly by all treatments compared to control plants. Treatment A increased the number of pods from 12.1 to 17.9 ($p < 0.001$), Treatment B to 19.0 and Treatment C increased the average number of pods to 17.6. The decreased number of flowers and concomitant increase in the number of pods per mainstem caused an overall increase in percentage pod set due to plant growth substance application (Table 7.4). The application of EL500 at growth stage 09 increased percentage pod set ($p < 0.001$), from 18.9% in control plants to 27.5%. The application of BAP + CLIAA at growth stage 41, increased percentage pod set to 34.5%. This also represented an increase ($p < 0.001$) over the single EL500 application. The addition of fertilizer, slightly decreased percentage pod set to 33.0%, which was a slight decrease compared to the plants treated, but without fertilizer (Treatment B). However, it also represented a significant increase ($p < 0.001$), over control plants (Table 7.4).

Effect of Density

Density had a significant effect on the number of flowers established on the mainstem. As density increased, the number of flowers decreased, from 72.8 at 10 plants m^{-2} , to 50.3 ($p < 0.001$), flowers at 40 plants m^{-2} . In general, as the density

Table 7.4: Effect of EL500, BAP, CLIAA, Fertilizer and Density on number of flowers, pod set, harvestable pods and pod retention.

Treatment	Density	No. Flowers	No. Pods Set	% Pod Set	No. Harvest Pods	% Harvest Pods	% Pod Retention
Control	10	72.2	13.9	19.2	9.0	12.8	66.8
	15	70.5	14.9	21.2	9.5	13.7	67.6
	20	65.8	13.6	19.9	7.5	11.3	54.2
	25	56.3	10.4	18.0	5.3	9.4	47.0
	30	59.1	11.5	19.9	7.1	12.1	63.0
	40	53.1	8.4	14.9	4.6	8.5	67.1
<i>Mean</i>		62.8	12.1	18.9	7.2	11.3	61.0
A	10	87.5	24.1	27.1	11.8	13.6	52.7
	15	71.5	18.7	28.5	9.7	14.5	54.3
	20	79.4	18.4	25.4	9.5	13.5	53.5
	25	66.8	17.1	27.6	9.4	15.0	55.3
	30	52.1	13.3	27.6	7.3	15.5	56.0
	40	54.0	15.7	28.6	7.5	14.8	55.6
<i>Mean</i>		68.6	17.9	27.5	9.2	14.5	54.6
B	10	65.3	21.8	33.3	11.9	19.2	62.9
	15	60.5	19.7	33.0	10.1	17.1	52.1
	20	63.8	20.7	33.0	11.1	18.1	57.0
	25	56.5	17.2	32.7	8.3	15.8	48.3
	30	49.1	16.5	34.4	8.5	17.9	53.1
	40	46.7	18.4	40.7	8.7	19.9	49.6
<i>Mean</i>		57.0	19.0	34.5	9.8	18.0	53.8
C	10	66.0	19.8	31.0	11.7	19.2	62.2
	15	62.6	21.3	34.7	10.9	18.3	52.4
	20	53.5	16.2	32.7	9.0	19.5	61.0
	25	55.5	13.9	25.6	7.4	13.7	53.2
	30	48.1	16.3	34.1	8.2	17.4	54.7
	40	47.6	17.9	39.8	8.0	18.5	46.6
<i>Mean</i>		55.5	17.6	33.0	9.2	17.8	55.0
Density Mean	10	72.8	19.9	27.7	11.1	16.2	61.1
	15	66.3	18.7	29.3	10.1	15.9	56.6
	20	65.6	17.2	27.8	9.3	15.6	56.4
	25	58.8	14.7	26.0	7.6	13.5	51.0
	30	52.1	14.4	29.0	7.8	15.7	56.7
	40	50.3	15.1	31.0	7.2	15.4	54.7
SED							
PGS		3.01	1.00	1.43	0.50	0.90	2.51
Density		3.69	1.23	1.75	0.61	1.10	3.07
PGS x Density		7.37	2.47	3.50	1.23	2.20	6.14
f value							
PGS (90df)		<0.001	<0.001	<0.001	<0.001	<0.001	0.018
Density (60df)		<0.001	<0.001	0.091	<0.001	0.168	0.045
PGS x Density (15df)		0.703	0.395	0.144	0.569	0.423	0.062

DATA SHOWN ARE THE MEAN OF 15 PLANTS

Table 7.5: Effect of EL500, BAP, CLIAA, and Plant Density on Harvestable Pods

increased, the number of pods set on the mainstem decreased. However, at 40 plants m⁻², the number of pods set increased slightly to 15.1 compared to 14.4 at a density of 30 plants m⁻². As with the number of flowers, the differences between adjacent densities was not significant. Both the number of flowers and pod set per mainstem decreased as the plant density increased, thus, the effect on percentage pod set was not significant. No significant effects were apparent between PGS treatment and density with respect to either number of flowers, pod set or percentage pod set (Table 7.4).

Intra-Raceme Percentage Harvestable Pods

Effect of PGS

The majority of harvestable pods on control plants were situated on flower position 1. The average percentage of flowers that produced harvestable pods at the proximal three flowers was 17.9%, while the average at the distal three flower positions was 0.3%. Application of EL500 at growth stage 09 increased the percentage of harvestable pods at flower positions 2 ($p < 0.001$) and 3 ($p < 0.01$), compared to control plants, resulting in an average percentage of harvestable pods of 22.7% on the proximal three flowers. No difference from control plants was observed at the distal three flowers (Table 7.5).

Application of BAP and CLIAA at growth stage 41 (Treatment B), further increased the percentage of flowers that had harvestable pods. Significant increases ($p < 0.001$), were observed at flower positions 2 and 3 compared to control plants, resulting in an average for the three proximal flowers of 28.0%. These increases at positions 2 and 3 were also significant ($p < 0.01$), compared Treatment A (Table 7.5). Average percentage of harvestable pods at the distal three flower positions was 1.0%.

Treat	Fls	Pods	Pod %	Fls	Pods	Pod %
PGS	40	14.4	17.9	40	15.1	17.9
Density	30	14.4	17.9	40	15.1	17.9
Plant Density	30	14.4	17.9	40	15.1	17.9

Table 7.5: Effect of EL500, BAP, CLIAA, Fertilizer and Density on intra-raceme percentage harvestable pods.

Treatment	Density (m ⁻²)	Flower Position					
		1	2	3	4	5	6
Control	10	38.7	19.3	2.0	0.4	0.0	0.0
	15	47.4	15.6	2.2	2.6	0.0	0.0
	20	37.7	16.3	1.1	0.0	0.0	0.0
	25	37.0	6.4	0.0	0.0	0.0	0.0
	30	40.4	13.4	4.6	1.3	0.0	0.0
	40	31.7	7.5	0.6	1.0	0.0	0.0
Mean		38.8	13.1	1.8	0.9	0.0	0.0
A	10	41.7	23.3	5.1	1.7	0.0	0.0
	15	36.9	23.0	7.1	1.2	0.0	0.0
	20	45.0	19.5	3.7	0.4	0.6	0.0
	25	42.2	22.9	6.9	0.0	0.0	0.0
	30	40.7	22.5	4.7	3.3	0.0	0.0
	40	40.0	18.4	4.7	1.1	0.0	0.0
Mean		41.1	21.6	5.3	1.3	0.0	0.0
B	10	43.7	37.0	13.2	4.0	0.0	0.0
	15	43.0	25.0	9.6	2.9	0.0	0.0
	20	44.7	33.1	4.0	6.9	0.0	0.0
	25	39.6	31.5	7.8	0.4	0.0	0.0
	30	43.6	32.5	5.9	0.8	0.0	0.0
	40	49.4	31.9	9.0	2.8	0.0	0.0
Mean		44.0	31.9	8.2	3.0	0.0	0.0
C	10	47.1	32.2	14.6	6.0	0.7	0.0
	15	43.8	31.2	12.5	0.7	0.0	0.0
	20	46.6	26.5	7.8	4.0	0.0	0.0
	25	41.0	25.0	2.2	0.6	0.0	0.0
	30	47.8	25.5	7.9	1.4	0.0	0.0
	40	49.3	27.4	7.6	3.1	0.0	0.0
Mean		45.9	28.0	8.8	2.6	0.1	0.0
Density Mean	10	42.8	28.0	8.7	3.0	0.2	0.0
	15	42.7	23.7	7.9	1.9	0.0	0.0
	20	43.5	23.8	4.2	2.8	0.1	0.0
	25	40.0	21.5	4.2	0.3	0.0	0.0
	30	43.1	23.5	5.8	1.7	0.0	0.0
	40	42.6	21.3	5.4	2.0	0.0	0.0
SED							
PGS		2.39	2.16	1.31	0.80	0.10	
Density		2.93	2.64	1.61	0.98	0.13	
PGS x Density		5.86	5.29	3.22	1.97	0.27	
f value							
PGS (90 df)		0.017	<0.001	<0.001	0.025	0.542	
Density (60 df)		0.868	0.142	0.018	0.075	0.526	
PGS x Density (15 df)		0.567	0.727	0.253	0.113	0.417	

Treatment C resulted in the greatest number of flowers at position 1 retaining harvestable pods, however, this increase was not significant. Increases at flower positions 2 and 3 ($p < 0.001$), compared to control plants, resulted in the average for the proximal three flowers being 27.6%. Increases were also observed compared to Treatment A ($p < 0.01$), but was not significantly different to Treatment B. Average percentage of harvestable pods on the distal three flower positions was slightly increased compared to control plants, to 0.9% (Table 7.5).

Effect of Density

Density had no effect on the percentage of flowers that retained harvestable pods, apart from at flower position 3. Plants grown at a density of 10 plants m^{-2} , retained the greatest percentage of harvestable pods (8.7%), while plants grown at densities of 20 and 25 plants m^{-2} had only 4.2% of flowers retaining harvestable pods ($p < 0.01$). No interaction between PGS and density was observed (Table 7.5).

Inter-Raceme Percentage Harvestable Pods

Effect of PGS

Control plants retained the majority of the harvestable pods on the lower racemes. Average percentage harvestable pods on the lower four racemes was 19.7%, on the upper four racemes it was 7.3% (Table 7.6)

Application of EL500 increased ($p < 0.01$), the percentage of harvestable pods at all four lower racemes, resulting in the average for the lower four racemes being 28.3% (Table 7.6). Increases were also evident at all four upper racemes, with a significant increase ($p < 0.05$), at raceme 5. Average percentage of harvestable pods on the upper five racemes was 9.8%.

Treatment B further increased the percentage of harvestable pods, compared to control plants ($p < 0.001$), at the lower four racemes, resulting in the average for

Table 7.6: Effect of EL500, BAP, CLIAA, Fertilizer and Density on inter-raceme percentage harvestable pods.

Treatment	Density (m ⁻²)	Raceme Number							
		1	2	3	4	5	6	7	8
Control	10	34.5	27.9	18.5	12.0	9.0	8.4	5.9	9.1
	15	36.0	25.5	16.5	13.6	12.3	12.6	8.5	8.3
	20	28.8	20.8	20.0	9.6	16.0	10.1	4.4	5.9
	25	20.2	14.3	14.3	9.6	9.5	9.4	5.1	1.3
	30	25.4	22.6	15.5	16.1	11.3	6.4	8.3	4.5
	40	29.8	17.8	13.4	9.5	4.5	1.7	1.0	1.0
Mean		29.1	21.5	16.4	11.7	10.4	8.1	5.5	5.0
A	10	40.0	32.8	26.6	21.3	19.2	14.6	12.2	6.7
	15	43.7	36.1	23.5	20.1	17.2	10.5	3.0	9.6
	20	31.9	36.8	23.9	14.9	10.5	8.4	7.6	7.1
	25	32.8	32.7	25.8	19.5	13.7	6.7	9.1	8.3
	30	42.1	34.2	22.2	14.4	13.9	7.2	3.7	4.4
	40	34.6	31.1	23.9	13.1	13.3	10.5	3.9	14.4
Mean		37.5	34.0	24.3	17.2	14.6	9.6	6.6	8.4
B	10	52.2	49.0	29.3	30.0	23.9	20.1	12.8	7.3
	15	49.6	34.8	29.6	21.4	15.5	11.7	9.0	7.7
	20	41.9	35.8	36.4	26.7	22.0	14.2	5.6	7.5
	25	45.5	39.8	27.9	18.5	13.5	5.8	3.5	1.0
	30	52.6	40.9	26.5	18.8	12.3	5.1	6.5	4.7
	40	41.3	37.1	28.1	22.3	15.0	9.8	6.9	2.0
Mean		47.2	39.6	29.6	23.0	17.0	11.1	7.4	5.0
C	10	53.8	50.8	30.2	26.6	17.1	20.6	7.3	5.4
	15	44.3	39.7	31.7	26.2	16.2	13.6	8.3	5.0
	20	41.1	42.1	26.0	21.3	17.5	10.8	5.1	3.5
	25	32.7	37.1	21.4	15.7	7.9	5.2	2.2	6.7
	30	39.5	32.9	30.3	19.5	22.5	5.1	1.6	2.4
	40	46.1	31.9	23.3	18.5	14.8	8.1	4.1	2.7
Mean		42.9	39.1	27.2	21.3	16.0	10.6	4.8	4.3
Density Mean	10	45.1	40.1	26.1	22.5	17.3	15.9	9.5	7.1
	15	43.4	34.0	25.3	20.3	15.3	12.1	7.2	7.7
	20	35.9	33.9	26.6	18.1	16.5	10.9	5.7	6.0
	25	32.8	31.0	22.3	15.8	11.1	6.8	5.0	4.3
	30	39.9	32.7	23.6	17.2	15.0	5.9	5.0	4.0
	40	37.9	29.5	22.2	15.9	11.9	7.5	4.0	5.0
SED									
PGS		3.01	2.38	1.87	1.73	1.97	1.75	1.57	1.85
Density		3.69	2.92	2.29	2.11	2.41	2.14	1.92	2.27
PGS x Density		7.38	5.83	4.58	4.23	4.82	4.29	3.85	4.54
f value									
PGS (90df)		<0.001	<0.001	<0.001	<0.001	0.005	0.336	0.359	0.109
Density (60df)		0.009	0.008	0.221	0.009	0.064	<0.001	0.055	0.503
PGS x Density (15df)		0.872	0.466	0.712	0.306	0.205	0.346	0.447	0.627

the lower four racemes being 34.9%. The average for the upper four racemes was increased to 10.1%. Increases were also evident at racemes 2 ($p < 0.05$) and racemes 1,3 and 4 ($p < 0.01$), over Treatment A (Table 7.6). Treatment C, resulted in a reduced percentage of harvestable pods at all racemes compared to Treatment B, however increases were evident ($p < 0.001$) over control plants at racemes 1 - 5. The average percentage of harvestable pods at the first four racemes was 32.6%, while on the upper four racemes it was 8.9%. (Table 7.6)

Effect of Density

Generally, on plants grown at all densities, the greatest percentage of harvestable pods was at the lower racemes. In addition, as the density was increased, the percentage of harvestable pods on any individual raceme was decreased (Table 7.6). Significant differences were observed between plants grown at 10 plant m^{-2} ($p < 0.01$) and those grown at a density of 25 and 40 plants m^{-2} at racemes 1,2,4 and 5. There was no interaction between the PGS treatment and density.

Overall Harvestable Pods

Effect of PGS

The number of harvestable pods retained on the mainstem of control plants was 7.2. Treatment A increased ($p < 0.001$), this number to 9.2, which in turn meant that the percentage of harvestable pods was increased from 11.3% to 14.5% (Table 7.4). Treatment B further increased ($p < 0.001$), the number of harvestable pods on the mainstem to 9.8. Percentage of harvestable pods was also increased by 6.7% ($p < 0.001$), compared to control plants (Table 7.4). Increases in both the number of pods and percentage of harvestable pods ($p < 0.001$), was also evident over Treatment A. Treatment C resulted in the same number of harvestable pods per mainstem as on plants treated with EL500 at growth stage 09 (Treatment A), however, due to the plants producing fewer flowers, the percentage of flower sites that produced harvestable pods was increased to 17.8%.

This was significantly higher ($p < 0.001$), than in control plants and Treatment A, but slightly less than Treatment B (Table 7.4).

Effect of Density

As the density of plants was increased, the number of harvestable pods per mainstem was decreased. Due to the number of flowers per mainstem also decreasing as density increased, density had no significant effect on the percentage of flowers that produced harvestable pods. No interaction was evident between PGS treatment and density with respect to either the number of harvestable pods or the percentage of flowers that produced harvestable pods (Table 7.4).

Plant Height

Effect of PGS

PGS treatments had no effect on the vegetative height of the plants (Table 7.7). However, the application of growth regulators, reduced reproductive plant height from 102cm in control plants, by up to 22% ($p < 0.01$).

Effect of Density

Vegetative height was in general, increased as density increased. Thus vegetative plant height was increased ($p < 0.05$), from 19.7 cm in plants grown at a density of 10 plants⁻² to 23.4cm in plants grown at 40 plants m⁻². Density had no significant effect on reproductive height (Table 7.7).

Table 7.7: Effect of EL500, BAP, CLIAA, Fertilizer and Density on vegetative and reproductive height.

Treatment	Density	Vegetative Height (cm)	Reproductive Height (cm)
Control	10	15.5	74.2
	15	18.9	107.3
	20	18.0	93.8
	25	20.7	119.3
	30	21.5	109.4
	40	24.0	110.0
	<i>Mean</i>	<i>19.8</i>	<i>102.3</i>
A	10	16.4	84.1
	15	17.6	82.6
	20	19.6	90.4
	25	20.1	86.9
	30	17.2	65.6
	40	24.1	93.1
	<i>Mean</i>	<i>19.2</i>	<i>83.8</i>
B	10	18.7	85.6
	15	17.6	86.4
	20	18.5	81.3
	25	19.9	75.8
	30	18.4	79.3
	40	24.5	76.6
	<i>Mean</i>	<i>19.6</i>	<i>80.8</i>
C	10	26.9	120.6
	15	16.9	81.9
	20	18.1	72.3
	25	20.1	95.9
	30	20.7	82.4
	40	21.1	87.8
	<i>Mean</i>	<i>20.6</i>	<i>90.1</i>
Density Mean	10	19.4	91.1
	15	17.7	89.6
	20	18.5	84.5
	25	20.2	94.5
	30	19.4	84.2
	40	23.4	91.9
SED			
PGS		1.49	7.15
Density		1.83	8.76
PGS x Density		3.65	17.51
f value			
PGS (90 df)		0.799	0.015
Density (60 df)		0.041	0.812
PGS x Density (15 df)		0.482	0.218

Mainstem Yield Analysis

Weight of Harvestable Pods

Effect of PGS

Control plants had on average 7.2 harvestable pods on the mainstem, which in turn had a fresh weight of 162.4g (Table 7.8). The application of EL500 at growth stage 09 (Treatment A), resulted in the weight of pods being increased ($p < 0.01$), by 23. % to 199.3g. Treatment B also increased fresh weight of pods by 17% ($p < 0.05$). The addition of fertilizer (Treatment C), also increased fresh weight of pods compared to control plants (14%, $p < 0.05$), however, this increase was slightly less than Treatment B (Table 7.8).

Effect of Density

In general, as the density was increased, the weight of harvestable pods on the mainstem was reduced, thus following the same pattern as for the number of harvestable pods. The only exception to this rule was once again the 25 plants m^{-2} density, which had a reduced weight of pods compared to 30 plants m^{-2} . Plants grown at a density of 40 plants m^{-2} , had a 44% ($p < 0.001$), reduction in the weight of pods on the mainstem, compared to plants grown at a density of 10 m^{-2} . There was no interaction between PGS application and density (Table 7.8).

Fresh Weight of Seeds

Effect of PGS

Fresh weight of seeds followed a similar pattern to that of fresh weight of pods, thus suggesting that all the pods had a similar number of seeds. Control plants had on average 43.7g of fresh beans on each mainstem. Treatment A, increased this by 32%, to 57.9g ($p < 0.001$). Increases compared to control plants due to treatments B and C, were 22% ($p < 0.05$) and 16% respectively (Table 7.8).

Table 7.8: Effect of EL500, BAP, CLIAA, Fertilizer and Density on the mainstem yield.

Treatment	Density	No. Harvest Pods	Wt. Harvest Pods (g)	Fresh Weight Seeds (g)	Dry Weight Seeds (g)
Control	10	9.0	197.9	64.1	18.8
	15	9.5	214.1	59.1	15.5
	20	7.5	157.5	41.3	10.7
	25	5.3	111.1	27.7	7.3
	30	7.1	172.7	42.1	12.1
	40	4.6	121.2	28.0	7.4
	<i>Mean</i>	7.2	162.4	43.7	12.0
A	10	11.8	243.0	64.8	17.0
	15	9.7	236.7	76.1	22.2
	20	9.5	212.8	63.4	18.4
	25	9.4	188.4	49.6	13.1
	30	7.3	149.3	43.6	12.9
	40	7.5	165.5	49.8	14.2
	<i>Mean</i>	9.2	199.3	57.9	16.3
B	10	11.9	245.2	66.8	18.0
	15	10.1	180.8	44.9	11.5
	20	11.1	207.6	58.0	16.1
	25	8.3	169.1	48.6	14.0
	30	8.5	164.8	47.8	13.3
	40	8.7	175.3	53.3	14.9
	<i>Mean</i>	9.8	190.5	53.2	14.6
C	10	11.7	208.5	59.7	17.8
	15	10.9	211.8	58.1	15.8
	20	9.0	199.7	56.3	15.9
	25	7.4	162.3	43.0	11.5
	30	8.2	169.4	45.3	12.8
	40	8.0	158.7	40.3	10.5
	<i>Mean</i>	9.2	185.1	50.5	14.0
Density Mean	10	11.1	223.7	63.9	17.9
	15	10.1	210.8	59.6	16.2
	20	9.3	194.4	54.8	15.3
	25	7.6	157.7	42.2	11.5
	30	7.8	164.1	44.7	12.7
	40	7.2	155.2	42.8	11.7
	<i>Mean</i>				
SED					
PGS		0.50	12.06	3.77	1.08
Density		0.61	14.77	4.62	1.32
PGS x Density		1.23	29.54	9.24	2.64
f value					
PGS (90 df)		<0.001	0.018	0.002	0.001
Density (60 df)		<0.001	<0.001	<0.001	<0.001
PGS x Density (15 df)		0.569	0.541	0.172	0.025

Effect of Density

As with the weight of harvestable pods, in general, the fresh weight of seeds on the mainstem, decreased as the density increased. 25 plants m^{-2} was the only exception to this rule with those plants having the lowest weight of seeds. Plants grown at a density of 10 plants m^{-2} , had 51 % ($p < 0.001$), more fresh weight of seeds compared to plants grown at 40 plants m^{-2} (Table 7.8). There was no interaction between the PGS application and density.

Dry Weight of Seeds

Effect of PGS

On average, control plants had a dry weight of 12g of seeds contained on the mainstem. This was increased by 36 % ($p < 0.001$), by the application of EL500 at growth stage 09. The application of BAP and CLIAA at growth stage 41, increased the dry weight of seeds compared to control plants by 22 %, whereas the same treatment, but with the addition of fertilizer (Treatment C), only increased it by 17 % (Table 7.8).

Effect of Density

The dry weight of seeds contained on the mainstem decreased with increase density from 17.9g at a density of 10 plants m^{-2} to 11.7g at 40 plants m^{-2} (53 %, $p < 0.001$). Plants grown at a density of 25 m^{-2} , again had the lowest weight of seeds. A slight trend of increased dry weight of seeds was evident as the plant population and number of treatments increased (Table 7.8).

Tiller Yield Analysis

Number of Tillers

Control plants had on average 1.64 tillers, this was increased ($p < 0.05$), to 2.1 tillers by the application of EL500 at growth stage 09 (Table 7.9). Other PGS treatments had no significant effect on the number of tillers.

As the density of the plants increased from 10 plants m^{-2} to 40 plants m^{-2} , the number of tillers per plant was reduced from 3.18 to 0.95 ($p < 0.001$). In addition, as the number of treatments applied and density increased, the number of tillers was reduced.

Number of Harvestable Pods

The application of plant growth substances to the plants had no significant effect on the number of harvestable pods contained on the tillers of the plants (Table 7.9).

As the plant density increased, the number of pods contained on the tillers was reduced. Plants grown at a density of 10 plants m^{-2} , had on average 10.3 harvestable pods on the tillers, whereas, plants grown at a density of 40 plants m^{-2} , had only 2.5 harvestable pods ($p < 0.001$). Plants grown at a density of 20 m^{-2} , did not fit into this pattern as these plants had fewer pods on than plants grown at 25 plants m^{-2} (Table 7.9).

Weight of Harvestable Pods

Application of PGSs had no effect on the weight of pods on tillers. Weight of harvestable pods was significantly altered by density, due to the significant effect on the number of tillers. Thus, as the density of plant population was increased, the weight of harvestable pods was reduced (Table 7.9). There was no relationship between density and plant growth substance application.

Fresh Weight of Seeds

The application of plant growth substances had no effect on the fresh weight of seeds of tillers. As plant population was increased from 10 plants m^{-2} to 40 plants m^{-2} , the fresh weight of seeds was reduced by 340% ($p < 0.001$). No relationship existed between the density and plant growth substance application (Table 7.9).

Table 7.9: Effect of EL500, BAP, CLIAA, Fertilizer and Density on the yield of tillers.

Treatment	Density	No. Tillers	No. Harvest Pods	Wt Harvest Pods (g)	Fresh Weight Seeds (g)	Dry Weight Seeds (g)
Control	10	2.07	7.1	131.1	35.9	8.6
	15	2.27	7.1	158.4	40.0	10.4
	20	2.00	5.0	96.7	24.8	6.2
	25	1.87	4.9	89.7	22.8	6.1
	30	0.80	2.8	51.0	12.3	3.3
	40	0.87	2.1	51.5	12.5	3.3
Mean		1.64	4.8	96.4	24.7	6.3
A	10	4.53	14.2	214.0	53.9	12.8
	15	2.27	6.3	124.4	33.5	8.7
	20	1.73	4.8	96.0	29.0	7.6
	25	2.00	7.3	147.5	38.1	10.2
	30	1.00	1.5	29.3	8.4	2.5
	40	1.07	2.0	44.3	10.6	2.5
Mean		2.10	6.0	109.2	28.9	7.4
B	10	3.20	10.1	184.7	48.1	12.0
	15	2.53	8.2	125.0	28.6	6.6
	20	1.93	7.1	118.3	32.0	8.2
	25	2.00	7.2	129.5	37.7	10.5
	30	1.00	4.3	75.2	20.4	5.4
	40	0.93	2.3	36.7	10.9	2.8
Mean		1.93	6.5	111.6	29.6	7.6
C	10	2.93	9.6	138.6	39.1	10.1
	15	2.53	8.1	167.1	42.2	10.7
	20	1.87	6.4	115.5	27.2	6.7
	25	1.53	5.0	107.6	28.4	7.4
	30	1.33	5.3	109.3	26.3	6.6
	40	0.93	3.7	72.3	17.6	4.7
Mean		1.86	6.4	118.4	30.1	7.7
Density Mean	10	3.18	10.3	167.1	44.2	10.9
	15	2.40	7.4	143.7	36.1	9.1
	20	1.88	5.8	106.6	28.2	7.2
	25	1.85	6.1	118.6	31.8	8.5
	30	1.03	3.5	66.2	16.9	4.5
	40	0.95	2.5	51.2	12.9	3.3
SED						
PGS		0.165	0.73	14.66	3.92	1.02
Density		0.202	0.90	17.96	4.81	1.25
PGS x Density		0.403	1.79	35.91	9.61	2.50
f value						
PGS (90df)		0.050	0.088	0.502	0.499	0.499
Density (60df)		<0.001	<0.001	<0.001	<0.001	<0.001
PGS x Density (15df)		0.003	0.092	0.308	0.552	0.518

Table 7.10: Effect of EL500, BAP, CLIAA, Fertilizer and Density on *mung bean*

Dry Weight of Seeds

Dry weight of seeds was reduced with increasing plant population, from 10.9g to 3.3g ($p < 0.001$). No effects were observed due to either plant growth substance application or interaction between PGS and density (Table 7.9).

Overall Yield Analysis

Number of Harvestable Pods

On average, control plants had 12 harvestable pods per plant. This was increased with the application of EL500 to 15.2 ($p < 0.01$). The application of BAP and CLIAA at growth stage 41 increased this figure to 16.3 (36% greater than control plants, $p < 0.001$). The application of fertilizer (Treatment C), resulted in a final number of harvestable pods of 15.5 (29% greater than control plants).

As the density of the plants increased, the average number of harvestable pods per plant was reduced. Plants grown at 10 plants m^{-2} , supported 220% ($p < 0.001$), more harvestable pods than those grown at a density of 40 plants m^{-2} (Table 7.10). No relationship existed between the application of plant growth substances and density.

Weight of Harvestable Pods

Application of plant growth substance had no effect on the weight of harvestable pods (Table 7.10). As the density of the plants increased, the weight of pods supported on these plants was reduced. Plants grown at a density of 10 plants m^{-2} , supported 180% ($p < 0.001$), greater weight of pods than those grown at a density of 40 plants m^{-2} . No relationship existed between application of plant growth substance and density.

Factor	EL500	BAP	CLIAA	Fertilizer
PGS (ANOVA)	<0.001	0.711	0.001	0.001
Density (ANOVA)	<0.001	<0.001	<0.001	0.001
PGS x Density (ANOVA)	0.126	0.397	0.534	0.349

Table 7.10: Effect of EL500, BAP, CLIAA, Fertilizer and Density on overall yield.

Treatment	Density	No. Harvest Pods	Weight Harvest Pods (g)	Fresh Weight Seeds (g)	Dry Weight Seeds (g)
Control	10	16.1	316.9	100.0	27.4
	15	16.5	358.3	99.1	25.9
	20	12.5	254.2	66.1	16.1
	25	10.1	200.9	50.5	13.4
	30	9.9	223.7	54.4	15.3
	40	6.7	172.6	40.5	10.7
	<i>Mean</i>	<i>12.0</i>	<i>254.4</i>	<i>68.4</i>	<i>18.1</i>
A	10	26.0	407.6	118.7	29.8
	15	16.0	261.0	109.6	30.2
	20	14.3	308.7	92.4	26.1
	25	16.7	335.9	87.6	23.3
	30	8.9	178.6	52.0	15.4
	40	9.5	209.8	60.4	16.7
	<i>Mean</i>	<i>15.2</i>	<i>300.2</i>	<i>86.8</i>	<i>23.6</i>
B	10	22.1	430.0	114.8	29.4
	15	18.3	305.7	73.5	18.1
	20	18.1	325.9	90.0	24.2
	25	15.5	298.6	86.4	24.4
	30	12.8	240.0	68.2	18.7
	40	11.0	212.0	64.2	17.7
	<i>Mean</i>	<i>16.3</i>	<i>302.0</i>	<i>82.9</i>	<i>22.1</i>
C	10	21.3	339.6	98.8	27.9
	15	19.0	378.9	100.4	26.5
	20	15.4	315.2	83.5	22.6
	25	12.4	269.9	71.4	18.9
	30	13.5	278.6	71.7	19.4
	40	11.7	231.0	57.9	15.3
	<i>Mean</i>	<i>15.5</i>	<i>302.2</i>	<i>80.6</i>	<i>21.8</i>
Density Mean	10	21.4	373.5	108.1	28.6
	15	17.5	351.0	95.7	25.2
	20	15.1	301.0	83.0	22.2
	25	13.7	276.3	74.0	20.0
	30	11.3	230.2	61.6	17.2
	40	9.7	206.4	55.8	15.1
	<i>Mean</i>	<i>15.5</i>	<i>302.2</i>	<i>80.6</i>	<i>21.8</i>
SED					
PGS		1.00	23.41	6.65	1.79
Density		1.24	28.68	8.14	2.20
PGS x Density		2.47	57.35	16.28	4.39
f value					
PGS (90df)		<0.001	0.111	0.038	0.020
Density (60df)		<0.001	<0.001	<0.001	<0.001
PGS x Density (15df)		0.156	0.597	0.534	0.349

Fresh Weight of Seeds

Control plants supported on average 68.4g of seeds. This was increased ($p < 0.01$), by 27% by the application of EL500 (Table 7.10). Treatment B and C increased the weight of seeds by 21 and 18% over control plants respectively.

As with number of harvestable pods, the fresh weight of seeds on the average plant, was reduced as the plant density increased. Plants grown at 10 plants m^{-2} on average had a 93% ($p < 0.001$), greater weight of seeds than those grown at 40 plants m^{-2} . No relationship existed between density and plant growth substance application.

Dry Weight of Seeds

Control plants had on average 18.1g of dry seeds. This was increased 30% ($p < 0.01$), by the application of EL500. Thus, the increase was similar to that in fresh weight of seeds. Similar increases to those achieved in fresh weight yields, were also achieved by treatments B and C, namely 22 and 20% respectively (Table 7.10).

As density increased, the total dry weight of seeds per plant decreased (Table 7.10). Plants grown at 10 plants m^{-2} , had 89% ($p < 0.001$), greater dry weight yield of seeds than those grown at 40 plants m^{-2} . No relationship existed between application of plant growth substances and density.

Distribution of Dry Matter

Effect of PGS

Control plants contained on average 50.8% of dry matter in the stem and leaves (Table 7.11). This was reduced with the application of EL500 by 34% ($p < 0.001$). Treatment B, reduced the amount of dry matter contained in the mainstem by 29% ($p < 0.001$), while Treatment C reduced mainstem dry matter by 37% ($p < 0.001$).

Table 7.11: Effect of EL500, BAP, CLIAA, and their combinations on the distribution of dry matter in the stem, leaf, pod hull and beans

The percentage of dry matter contained in the pod hull of control plants was 27.9% (Table 7.11). EL500 application increased this by 18% ($p < 0.01$). BAP + CLIAA increased the percentage of dry matter in the hull compared to control plants by 14% ($p < 0.01$) and with the addition of fertilizer by 15% ($p < 0.01$).

Of total mainstem dry matter, 23% was contained in the seeds of control plants. This was increased by 41% ($p < 0.001$) by the application of EL500. Treatment B and C increased this figure by 43 and 45% respectively (Table 7.11).

Effect of Density

No definite pattern emerged from the effect of density on the percentage of mainstem dry matter contained in the actual stem and leaves. Plants grown at densities of 10 and 20 plants m^{-2} resulted in the lowest percentages, with those grown at 25 plants m^{-2} , having a higher percentage ($p < 0.01$). A similar trend occurred with respect to the amount of dry matter contained in the pod hull, i.e. plants grown at 10 and 20 plants m^{-2} , resulted in the highest percentages, while those grown at 25 plants m^{-2} had the lowest percentage of dry matter in the pod hull. Density had no effect on the amount of dry matter contained within the beans (Table 7.11).

Effect of Density x PGS

As the density and number of applications of plant growth substance was increased, the percentage of dry matter contained in the stem and leaf was reduced. Contrary to this as the density and applications were increased, so the amount of dry matter contained in the pod hull and beans^{increased} (Table 7.11).

Table 7.11: Effect of EL500, BAP, CLIAA, Fertilizer and Density on the distribution of dry matter in the mainstem and overall harvest index.

Treatment	Density	% Dry Matter in Stem + Leaf	% Dry Matter in Pods	% Dry Matter in Seeds	Harvest Index
Control	10	40.0	28.5	31.5	24.1
	15	49.6	29.4	21.1	20.7
	20	42.9	33.1	24.0	22.0
	25	65.0	21.6	13.4	12.1
	30	52.7	28.3	19.1	16.9
	40	54.7	26.4	18.9	18.8
<i>Mean</i>		50.8	27.9	21.3	19.1
A	10	38.9	34.1	27.0	23.9
	15	33.3	30.5	36.2	32.9
	20	42.2	33.3	28.9	25.5
	25	40.5	34.9	24.7	23.9
	30	31.0	34.7	34.3	32.2
	40	40.3	30.4	29.3	25.2
<i>Mean</i>		37.7	33.0	30.1	27.3
B	10	37.4	33.9	28.7	26.2
	15	57.7	23.4	19.0	15.6
	20	36.0	33.1	30.9	28.3
	25	35.4	32.2	32.4	31.4
	30	36.2	38.1	32.6	27.7
	40	34.5	31.0	39.1	38.4
<i>Mean</i>		39.5	31.9	30.5	27.9
C	10	27.0	36.9	36.1	31.6
	15	34.9	32.0	33.1	28.0
	20	32.0	32.8	35.1	29.6
	25	42.9	30.3	26.9	25.4
	30	42.4	29.8	27.8	23.9
	40	43.5	30.3	26.2	24.3
<i>Mean</i>		37.1	32.0	30.9	27.1
Density Mean	10	35.9	33.3	30.8	26.5
	15	43.9	28.8	27.3	24.3
	20	38.3	33.1	29.7	26.3
	25	45.9	29.8	24.3	23.2
	30	40.6	32.7	28.5	25.2
	40	43.3	29.5	28.4	26.7
SED					
PGS		2.56	1.42	2.00	1.92
Density		3.14	1.74	2.45	2.35
PGS x Density		6.28	3.49	4.91	4.70
f value					
PGS (36df)		<0.001	0.002	<0.001	<0.001
Density (24df)		0.017	0.021	0.147	0.616
PGS x Density (6df)		<0.001	0.049	0.001	0.001

DATA ARE MEAN OF 6 PLANTS

Harvest Index

Control plants had on average a harvest index of 19.1%. The application of EL500 increased this to 27.3 ($p < 0.001$). Application of BAP and CLIAA increased the ratio to 27.9, while with the addition of fertilizer it was 27.1. Density had no effect on the harvest index, but it was found that as density was increased in conjunction with the addition of plant growth substances, the harvest index ratio increased (Table 7.11).

Discussion

As in previous experiments, it was found that the application of plant growth substances to *Vicia faba*, could increase both overall pod set, number of harvestable pods and the overall yield. In this experiment, it was found that the greatest increase in yield was produced by the application of EL500 at growth stage 09. The application of cytokinins in conjunction with EL500 caused a significant ($p < 0.001$) increase in percentage pod set over both control plants and the single application of EL500, however, this increase did not continue through until final yield.

It was found in Chapters 4 and 6, that the exogenous application of BAP could significantly increase pod set compared to control plants. In this experiment, these findings were supported, but the levels of pod set were not as great as in Chapter 4, but similar to those achieved in Chapter 6.

It was also discovered in Chapter 4, that the application of CLIAA, after pollination increased percentage pod set (Rylott and Smith, 1990). Application of CLIAA + BAP via a drop leg sprayer technique, in Chapter 6, was found to increase both pod set and pod retention. Overall percentage pod set was 45%, compared to 15% in control plants. In this experiment, the average for the same treatment was 35%

compared to 19%. Thus, in the field trial, control plants set a greater percentage of pods, while the treated plants did not set as many. As in Chapter 6, although the overall number of harvestable pods was not significantly increased due to the application of BAP + CLIAA at growth stage 41, the percentage of harvestable pods at raceme 4 and below was significantly increased ($p < 0.001$), compared to the treatment not receiving the second application of plant growth substance (Treatment A). As a consequence of this it can be stated that the application of BAP + CLIAA, later in the plants development increases pod retention.

It was suggested in Chapter 6 that the application of artificial fertilizer to increase the production of assimilates may aid pod retention and final yield. However, although pod retention was increased (compared to Treatment B), overall yield was reduced compared to Treatments A and B. This agrees with previous findings, where seed-bed applications or split dressings with later applications of nitrogen (125-300kg/ha), resulted in small or negative yield responses (Mc Ewen, 1970 a,b; Mc Ewen *et al.*, 1973; Mc Ewen *et al.*, 1981).

The application of fertilizer at growth stage 31, resulted in some leaf scorch and in consequence a reduction in photosynthetic leaf area. It was found in Chapter 5, that the assimilates produced by the leaves remains an important factor for pod set until the end of flowering. Thus, the reduction in pod set associated with the application of fertilizer, could be accounted for by this reduction in leaf area. During pod fill, it was discovered in Chapter 5, that the predominant factor leading to increased weight of pods, was a reduction in stem height and weight. Plants that had fertilizer applied at growth stage 31 had on average, a taller reproductive height than either of the other treatments. Thus, the reduction in yield associated with the application of fertilizer may be a result of excessive stem growth and consequent waste of assimilates. Similar effects were found by Smith (1982), where irrigation lead to increased vegetative growth and a reduction in pod set. The distribution of

dry matter within the mainstem, however, does not appear to support this theory as the percentage of total mainstem dry matter contained within the stem and leaves was no greater in Treatment C than in any other treatment.

It can be concluded that the application of fertilizer at growth stage 31, did not have the effect of increased yield of seeds that was anticipated in previous chapters. Part of the reason for this may be due to leaf scorch and thus insufficient fertilizer may have been applied to overcome this loss of photosynthetic leaf area.

It was found in this experiment, that as the density of the plants was increased from 10 plants m^{-2} to 40 plants m^{-2} , the percentage of flowers that set pods, in either control or treated plots, remained unaffected. This directly contradicts results (Smith, 1982), where increased density (7 - 22 plants m^{-2}), increased flower abscission, especially at middle and distal flower positions. The percentage of mainstem flowers that resulted in harvestable pods was also unaffected by density. However, as the density increased, the number of harvestable pods per mainstem was reduced, whether treated or not. Similar results have been described by Ishag (1973a) and Smith (1982). As a consequence of this, it was found that the yield of seeds was reduced on the mainstem as the density was increased. Once again, these findings agree with those of Ishag (1973a) and Smith (1982). In addition, due to the lower density plants forming more tillers (Hodgeson and Blackman, 1957), which were also able to support harvestable pods, the yield of seeds per plant was also reduced as the density of plants increased.

From calculations carried out to find the yield per unit area, it was found, that as density increased, yield m^{-2} was also increased. Similar results were shown by Soper (1952), Sprent *et al.* (1977), Keller and Burkhard (1981) and Smith (1982). In control plants yield m^{-2} increased from 1000g (10 plants m^{-2}), to 1620g (40 plants m^{-2}). Optimum plant population was 30 plants m^{-2} , which yielded 1632g m^{-2} .

In EL500 treated plants the increase followed a similar pattern, with 40 plants m^{-2} yielding 2416g m^{-2} compared to 1187g at 10 plants m^{-2} . In Treatment B, yields were 1148g m^{-2} and 2568g m^{-2} , while in Treatment C were 988g m^{-2} and 2316g m^{-2} . Thus, the optimum plant population for control plants in this experiment would appear to be 30 plants m^{-2} . However, when plant growth substances are applied, the optimum plant population was increased to 40 plants m^{-2} . Greatest yield per unit area was achieved by Treatment B at 40 plants m^{-2} , which represented an increase in yield of 57% over the optimum plant population in control plots. If 57% yield increase were transposed into financial terms, it is likely that the increased expenditure for seeds, chemical and workload would be well rewarded in financial return, so leading to an increased gross margin.

The application of a single dose of EL500 at growth stage 09 was made at a similar site in 1987 (Chapter 3). Plants were then grown at a density of 30 plants m^{-2} . Control plants in 1987, set 17.2% of pods, while treated plants set 42%. Plants grown at the same density in 1988 set 19.9% in control plants, and 27.6% in EL500 treated plants. It would appear from these results therefore, that the level of pod set in control plants remains similar, while those in treated plants is more variable. Yields of control plots in 1987 were 62.3g of seeds per plant compared to 87.7g of seeds in treated plants. In 1988, the yield of control plants grown at the same density was 54.4g while in EL500 treated plants was 52.0g. Thus, in 1987, the treated plants increased the yield of seeds, while in 1988 treated plants resulted in a reduction in the yield of seeds. Although 1988 yields were reduced in both control and treated plants, the reduction was more significant in the treated plants.

Climatic conditions at the time of spraying in both years were as follows: In 1987 the maximum temperature was 16.6°C, and the minimum was 8.5°C. No rainfall was recorded. In 1988, temperature ranged from a max. of 17.5°C to a min. of 13.7°C. Rainfall on the day of spraying was recorded as 10mm and began to fall 2

hours after the spray had been applied. It can be suggested therefore, that at the time of application and for the next few hours, the temperature was quite similar. However, in 1988, rainfall 2 hours after application may have reduced the uptake and efficiency of the EL500. The rainfastness of the chemical is unknown due to its current experimental status. However, a similar anti-gibberellin growth retardant (chlormequat) is not rainfast until 6 hours after application (Ivens, 1990). It would appear logical therefore, that any applications of growth regulator should include the addition of a sticker such as "Guard", which will render the chemical rainfast in 30-60 minutes (Heapy, personal communication). Similarly, the use of other adjuvants such as uptake enhancers may have increased the stability of the results.

It must also be stated, however, that the application of a single growth regulator to broad beans may not be sufficient to reduce the crops supposed yield instability, i.e. combinations such as those in Treatment B, may be necessary. Therefore, trials would need to be continued over a greater number of years before a definite stability index could be established.

Chapter 8

Chapter 8

Effect of hand-tripping and the application of BAP on parthenocarpic pod set

Introduction

It was established in Chapter 4 that application of the synthetic cytokinin, 6-benzylaminopurine (BAP), to flowers of broad beans dramatically increased pod set (Rylott and Smith, 1990). However, it was unclear from this experiment whether the observed increase in pod set was due to an increase in parthenocarpic pod set. The aim, therefore, of this experiment was to establish the inherent ability of the variety to set pods and the extent that this was affected by the application of BAP.

Method

Plants of the variety Threefold White were grown in a bee-proof glasshouse, under natural light conditions at the Edinburgh School of Agriculture during April to June 1989. All other methods of raising the plants were the same as in Chapter 4.

The experiment was arranged as a randomised block containing 5 replicates of the treatments (Table 8.1). BAP ($1 \times 10^{-4} \text{M}$) and the control, distilled water, were applied to the standard petal and calyx using a fine artists paint brush. Flowers were tripped 24 hours after treatment, where applicable. Flowers were judged to be at the correct stage for tripping or application of the growth regulator at flower development stage 9. Pod set was recorded when the pods were 1-2cm long. The pods were then removed from the plant, dissected and the number of embryos (if present), recorded. Presence of a fertilised embryo was recorded if the seed showed evidence of expansion when viewed under an eye lens ($\times 10$). If the seed site appeared as a small black point, the embryo was considered to have not grown and was recorded as not present.

Table 8.1: Description of Treatments

Treatment	Plant Growth Substance	Tripped/Untripped
A	Distilled Water + "Agral"	Untripped
B	Distilled Water + "Agral"	Tripped
C	6-benzylaminopurine	Untripped
D	6-benzylaminopurine	Tripped

Results

Intra-Raceme Percentage Pod Set

The pattern of pod set in both untripped and tripped control plants followed the usual pattern i.e. dominance of pod set at proximal flower positions on the raceme (Fig. 8.1a). Untripped flowers (Treatment A), set pods in 25 and 11% of cases at positions one and two respectively. Hand-tripping the flowers on control plants (Treatment B), resulted in increased pod set at all flower positions compared to the non-tripped plants. This increase was significant ($p < 0.01$), at flower positions one and two where pod set was 64 and 44% respectively.

The application of BAP to non-tripped plants (Treatment C), resulted in significant increases ($p < 0.001$), in pod set compared to non-tripped control plants at all flower positions apart from position 4 (Fig. 8.2a). BAP applied 24 hours before tripping (Treatment D), resulted in highly significant ($p < 0.001$), increases in pod set compared to all other treatments and at all flower positions (Figs. 8.3a, 8.4a).

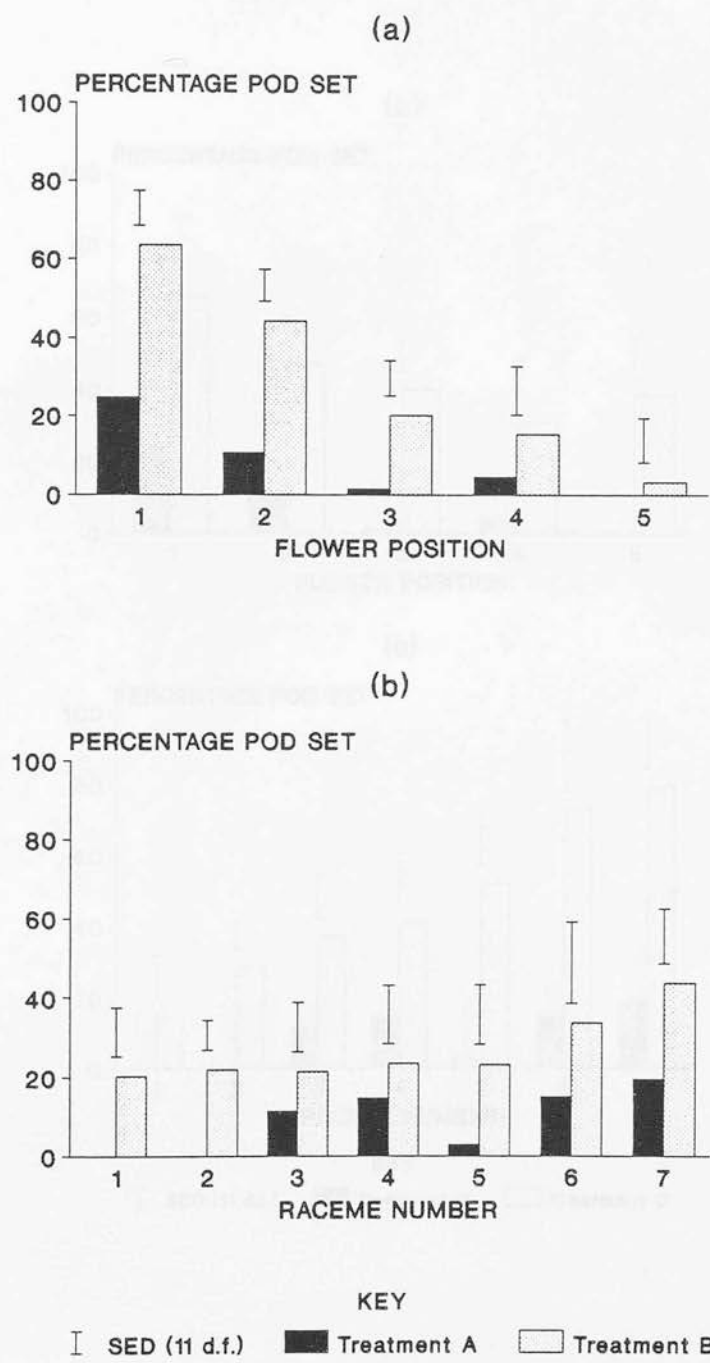


Figure 8.1: Effect of Hand-tripping on a) intra-raceme and b) inter-raceme pod set. (Actual figures are shown in Appendices 8.1 and 8.2).

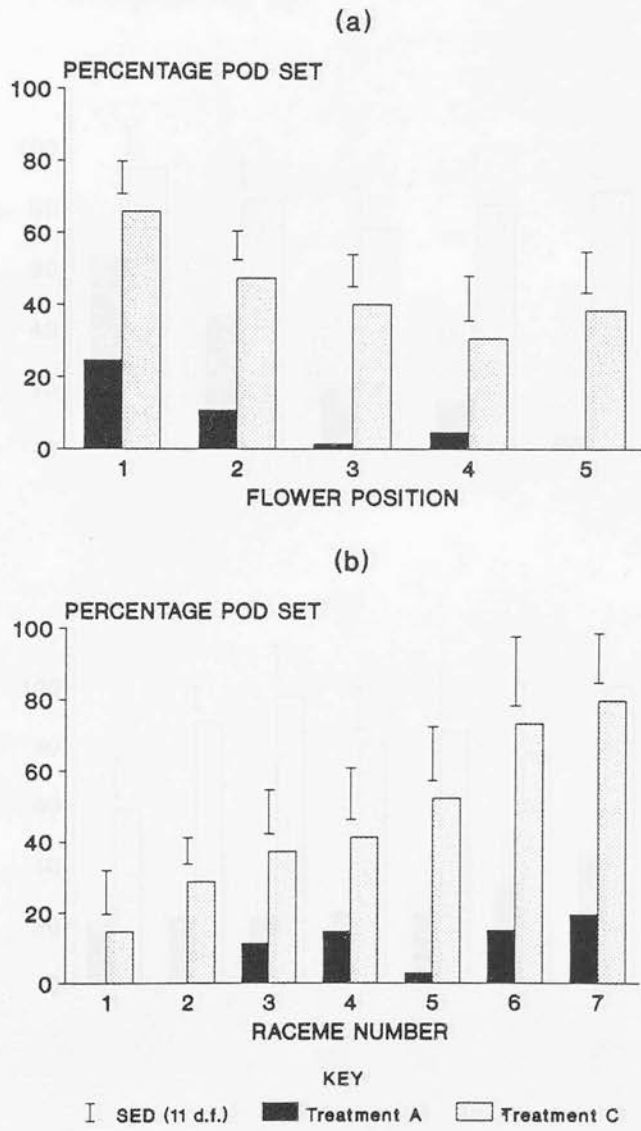


Figure 8.2: Effect of application of BAP on untripped plants on a) intra-raceme and b) inter-raceme pod set. (Actual figures are shown in Appendices 8.1 and 8.2).

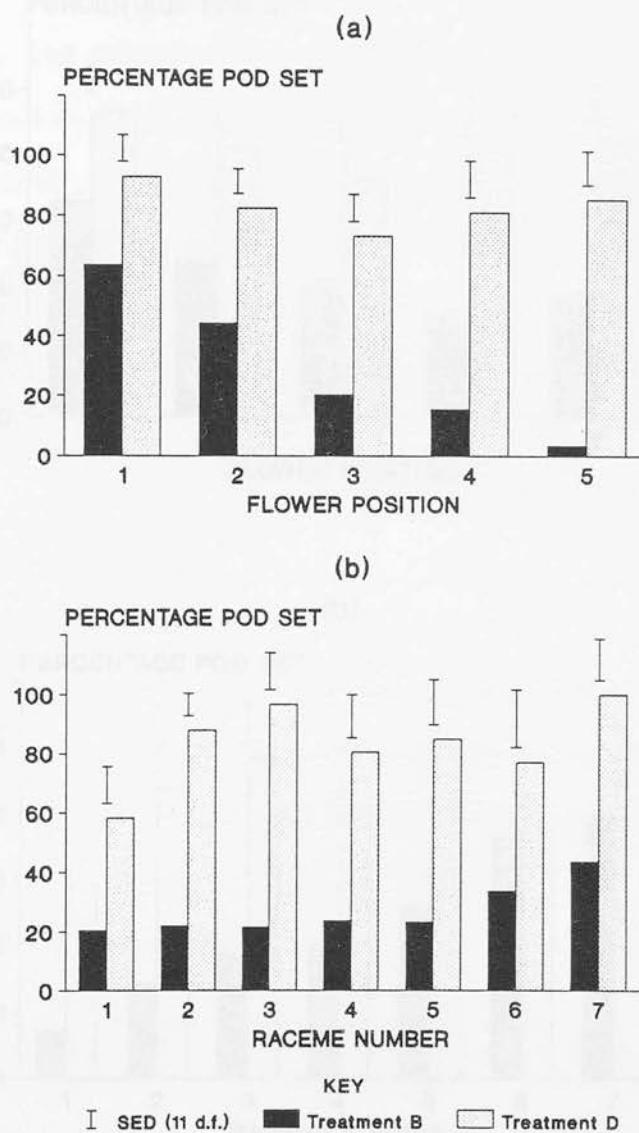


Figure 8.3: Effect of BAP on hand-tripped plants on a) intra-raceme and b) inter-raceme pod set. (Actual figures are shown in Appendices 8.1 and 8.2).

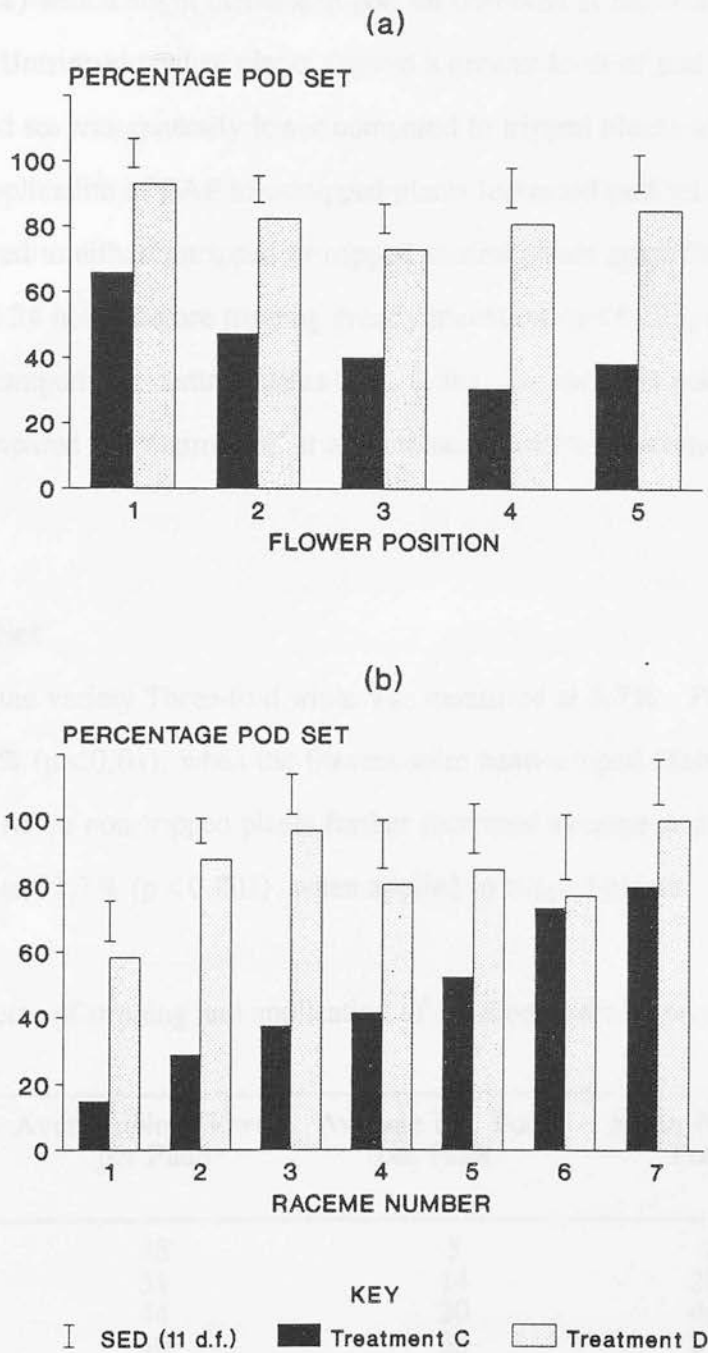


Figure 8.4: Effect of BAP applied to tripped and untripped flowers on a) intra-raceme and b) inter-raceme pod set. (Actual figures are shown in Appendices 8.1 and 8.2).

Inter-Raceme Percentage Pod Set

Hand-tripped control plants set similar numbers of pods on each of the first 5 racemes (21-24%) with a slight increase in pod set observed at racemes 6 and 7 (34 and 44%). Untripped control plants showed a greater level of pod set on upper racemes, but pod set was generally lower compared to tripped plants at all racemes (Fig. 8.1b). Application of BAP to untripped plants increased pod set at all racemes compared to either untripped or tripped control plants apart from at raceme 1. BAP applied 24 hours before tripping greatly increased ($p < 0.01$), pod set at all racemes when compared to control plants (Fig. 8.3b). Increases in pod set were also evident compared to Treatment C at all racemes apart from raceme 6 (Fig.8.4b).

Overall Pod Set

Autofertility of the variety Three-fold white was measured at 8.7%. Pod set was increased by 20% ($p < 0.01$), when the flowers were hand-tripped (Table 8.2). The application of BAP to non-tripped plants further increased average pod set to 46.7% ($p < 0.001$) and to 81.7% ($p < 0.001$), when applied to tripped plants.

Table 8.2 : Effects of tripping and application of BAP on overall pod set.

Treatment	Average No. Flowers per Plant	Average No. Pods per Plant	Mean Percentage Pod Set
A	48	5	8.7
B	51	14	28.7
C	44	20	46.7
D	40	33	81.7
SED (11 d.f.)	8.8	4.2	5.29
f value	0.629	<0.001	<0.001

Evidence Of Seed Development In Pods

All pods in all treatments tested for signs of seed development contained at least one seed that showed signs of expansion. Tripping and the application of BAP had no significant effects on the number of seeds per pod. All pods which set on untripped control plants (Treatment A) contained either 4 or 5 seeds per pod, with the majority (62%) containing 5 seeds (Table 8.3) . The other treatments all contained similar numbers of seeds per pod, i.e. 4 seeds (50 - 52%) and 5 seeds (35 - 38%).

Table 8.3: Effect of tripping and application of BAP on the number of expanded embryos per pod expressed as the proportion (%) of total pods on plant.

Treatment	% Pods With 1 Seed	% Pods With 2 Seeds	% Pods With 3 Seeds	% Pods With 4 Seeds	%Pods With 5 Seeds
A	0.0	0.0	0.0	37.8	62.2
B	1.2	1.4	6.3	52.9	38.2
C	0.7	4.0	8.4	50.4	36.5
D	0.0	2.9	5.9	55.4	35.9
SED (11 d.f)	0.99	1.99	3.75	19.52	19.46
f value	0.589	0.263	0.193	0.809	0.490

Discussion

The autofertility rate of the variety Three-fold White was measured as 8.7%. Published estimates of autofertility for *V. faba* range from 4 - 84% (Bond and Poulsen, 1983). Pod set on untripped control plants demonstrated the typical pattern, i.e. dominance at proximal positions within the raceme. This suggests that the first formed flower on each raceme is either more autofertile, or possibly the intrinsic hormonal concentrations at these flower positions are more conducive to an

extended flower attachment period. Either of these explanations would allow the flower a greater opportunity to self-fertilize. The inter-raceme pod set pattern on untripped control plants indicated that flowers on upper racemes were more autofertile. This agrees with previous findings by several authors, as discussed by Bond and Poulsen (1983).

Intra-raceme pod set of hand-tripped plants followed the usual pattern seen in all previous experiments. Inter-raceme pod set was, however, slightly different in that flowers on the upper racemes tended to set a greater percentage of pods. Normally this trend is reversed. The reason for this may be that although the plants flowered in May-June, due to the plants being grown in the glasshouse without supplementary lights, the lower racemes were short of assimilates. This may be due to the increased growth rate of plants in the glasshouse, combined with mutual shading of the plants especially at the lower racemes (Hodgeson and Blackman, 1957; Smith, 1982). Hence, the apex is in less competition for light and so continues to attract a greater percentage of the assimilates than lower racemes.

The pattern of pod set on the BAP treated tripped plants in this experiment closely followed those found in Chapter 4. From this it can be concluded that the application of exogenous cytokinin to the flower overrides the natural plant hormonal system (which normally results in proximal pod dominance). This allows all pods both within and over all racemes an equal opportunity of setting pods. Rylott and Smith (1990), suggested that the reason for this increase in pod set was due to the exogenously applied cytokinin being responsible for promoting vascular differentiation at the pedicle:peduncle junction by attracting assimilates to the developing sinks and stimulating cell division. This increased attraction of assimilates may also be why the BAP tripped plants did not show a tendency for greater pod set at the upper racemes, as in control plants.

Chapman *et al.*, (1979), stated that the removal of the stem apex resulted in both decreased flower abscission and increased parthenocarpic pod set. It may have followed therefore, that the application of BAP to the flowers and the resultant decrease in flower abscission may have been accompanied by an increase in parthenocarpic pod development. In this experiment, pods of variety Threefold White had an autofertility rate of 8.7%. These pods when dissected (3 - 6 days after treatment), were found to contain either 4 or 5 enlarged seeds. Hand-tripping resulted in greater pod set, but the number of pods containing at least one enlarged seed remained the same i.e. 100%. Of these pods, most contained either 4 or 5 seeds. The application of BAP to either tripped or untripped flowers resulted in significantly ($p < 0.001$) increased pod set. All of these pods, again, contained seeds and the number of seeds remained similar to that of control plants. It can therefore be concluded that the exogenous application of BAP did not induce parthenocarpic pod set. The primary effect of BAP was to increase pod set by delaying the abscission of reproductive structures thereby allowing a greater opportunity for the flower to self. In the field, this extended flower retention period would give the flower a greater opportunity of being cross-pollinated and as a consequence the yield of harvestable pods is likely to be greater.

Chapter 9

Discussion and Conclusions

Apert from the application of 100% of the recommended rate of 150 kg N/ha, all plant growth responses were significantly reduced and yield 2.43 t/ha. All plant growth responses were significantly reduced by 50% and 75% compared to the recommended rate of 150 kg N/ha, although plant growth was improved in some respects of some, but not all, attributes were not always shown. Differences were observed that were highly significant in control plant was 1.7 g - 13.4 g. The application of plant growth responses showed in much what a variation in nitrogen rate was plant growth in the relevant control plant ranged from 100% (24.1 g - 13.4 g) to 50%.

In Chapter 7, it was demonstrated that the yield of control plant grown in 1987 and 1988, was more variable in plants treated with a single application of 150 kg N/ha than the suggested 100 kg N/ha, and under a single application of 100 kg N/ha, a control plant showed, or that did not, a significant response over a greater number of years and also in 1987 and 1988. It must be stated that a coefficient of variation of 100% is not a good one. For example, a control plant grown in 1987 and 1988 should be tested against control plants in 1987 and 1988 and over a greater number of years (5 - 10).

Much of the variation in control plant yields (especially in the above experiments) can be ascribed to the greater variability present within the same level of the various levels of cross breeding during seed development. Different levels of cross breeding result in different levels of variability and therefore yield in yield comparison (Hewitt et al., 1980). In addition, the small number of replicates (5), may have compounded this problem. Indeed, in the field trials, where 15 plants

As discussed in Chapter 1, the major factor leading to instability and loss of yield potential, is abscission of flowers either pre or post pollination (Gates *et al.*, 1983b). In general, it was shown that the application of plant growth substances to *Vicia faba*, can reduce the amount of flower drop and so increase pod set in the crop. In control plants pod set in the different experiments varied between 10.9 - 18.9%. Apart from the application of PP333 at growth stage 03 (Chapter 3), which reduced pod set by 2.4%, all plant growth substance applications increased pod set by between 0.5 and 86.1%, compared to the control plants in the experiment. Although pod set was increased in the vast majority of cases, final yield increases were not always evident. Differences in the average fresh weight yield of seeds per control plant was 45.7g - 77.4g. The application of plant growth substances resulted in much greater variation, as average yield per plant compared to the relevant control plants ranged from -56% (34.1g), to + 52% (94.7g).

In Chapter 7, it was demonstrated that the yield of control plants between 1987 and 1988, was not as variable as plants treated with a single application of EL500. It must be suggested therefore, that either a single application was insufficient to sustain clear yield advantages, or that such applications must be tested over a greater number of years and climatic conditions, in order to establish a true coefficient of variability of yield. For example, a series of plant growth substance applications should be tested against control plants at different sites and over a greater number of years (5 - 10).

Much of the variation in control plant yields (especially in glasshouse experiments), can be ascribed to the genetic variability present within the crop due to the various levels of cross breeding during seed multiplication. Differing levels of cross-breeding results in differing levels of autofertility and inbreeding leads to yield depression (Lawes *et al.*, 1983). In addition, the small number of replications (5), may have compounded this problem. Indeed, in the field trials, where 15 plants

were harvested, variability in both pod set and yield of control plants was not as great. As discussed in Chapter 3, inbred lines would lead to reduced levels of variability, but the results obtained would not relate to commercial practice. It would appear, that in order to discover true effects, the number of replications may need to be increased to at least 15. Obviously, this in turn leads to greater problems of workload and so fewer growth regulator treatments could be screened at any one time.

Variability of results between those carried out in the glasshouse and those carried out in the field, or between those treatments cross-referenced between years can be partly explained by the above arguments about plant variability. However, it is more likely that much of this variability was due to the efficiency of the growth regulator at or directly after application. Several factors affect efficiency including: uptake, temperature, light, pH of the soil, and formulation of the active material (Keller and Belluci, 1983). Levels of intrinsic hormones vary with physiological age (Engvild, 1985) and these differing levels at the time of application can affect plant growth substance efficiency (Keller and Belluci, 1983). Thus, during this series of experiments, all applications were made at specific physiological growth stages (related to flower and pod development), in order to try and reduce some of this variability. Within the glasshouse experiments, conditions of temperature, water supply, light and soil conditions remained constant both during an individual experiment and between different experiments, thus allowing a direct comparison. In the field, however, differing climatic conditions at the time of application or during subsequent plant development may have led to differing responses (Chapter 7, discussion). It was shown by Lloyd-Jones (1973), that the penetration of daminozide into apple leaves over a 96 hour period could range from 10 to 80%. Foliar penetration, however, depends to a very large extent on cuticle structure (Luckwill, 1978), with this being dependent on a range of environmental factors including temperature under which the plant was grown and physiological age of the

plant structure (Berry, personal communication). In order to reduce some of these problems, it is suggested that in future, more emphasis be placed upon the climatic conditions suitable for chemical uptake and response. One possible solution would be to first screen a series of adjuvants e.g. wetters, stickers and uptake enhancers, which may increase chemical efficiency over a range of conditions by effectively improving the chemical formulation. The use of adjuvants such as LI700 has been shown to increase the uptake of chlormequat in cool conditions (Newman, personal communication).

Experiments (Chapter 4), revealed that the intrinsic hormonal control of pod set was probably related to levels of intrinsic cytokinin (pre-pollination) and/or auxin post-pollination (Rylott and Smith, 1990). The application of artificial plant growth retardants, altered plant development (Chapter 5), causing increased root and pod growth compared to decreased stem growth. It was concluded from this, that the application of plant growth retardants may increase the free cytokinin:gibberellin ratio, while leaving levels of auxin relatively unaffected and so allow increased pod set (Rylott and Smith, 1990). Based upon these findings, it can be suggested that future experimentation to increase the yield of faba bean could follow several important routes: 1. The screening of plant growth regulators in conjunction with assays to discover which artificial growth retardants most successfully increase the cytokinin levels within the plant. 2. The application of different natural products such as seaweed extracts that are known to be high in cytokinins (Thomas and Blakesley, 1987). 3. Breeding of plants for increased levels of cytokinins. Smith (1982), discovered that ideotypes with an independent vascular supply to flowers exhibited high levels of pod set. This system allows all flowers within the raceme to import similar amounts of hormones and assimilates. Although this argument cannot be discounted, it could be suggested that lines that appear to exhibit this characteristic, may also exhibit increased levels of intrinsic cytokinin.

From the experiments performed, it has been shown that it is possible to manipulate pod set in *Vicia faba* with plant growth substances. However, this increased yield potential is not always carried through to similar increases in overall yield (Chapters 3, 4, 6 and 7). It would appear therefore, that after initial pod set problems have been overcome hormonally, by the application of cytokinins pre-pollination to establish sink sites and auxins post-pollination to induce vascular differentiation, the next factor is one of increasing sink strength in the developing pods. Jaquiere and Keller (1978), stated that at the end of flowering, the apex passed on its role as principal sink to the developing pods. Thus, pods do not become active sinks until they are 4 - 6cm in length. Much of the increased yield potential (due to increased pod set, achieved by plant growth substance application), was lost during this critical stage between pod set (1 - 2cm in length) and the pods attaining active sink status, due to increased levels of pod drop.

Sink strength is measured by its absolute rate of increase in dry weight and is the product of its size and activity, which in turn is measured by the potential rate of metabolite uptake per unit weight of sink tissue per unit time (Wareing, 1978). The supply of assimilates (mainly sucrose), to the seed (the primary sink involved in the yield of faba beans), is delivered via the phloem to the apoplast, which separates the maternal tissue from that of the next generation (Hay and Walker, 1989). The rate at which assimilates are accumulated by a sink can therefore be limited either by the rate of unloading or by the uptake from the apoplast (Wareing, 1978). Where there is competition of assimilates between various sinks within the plant, the effectiveness of that sink to accumulate assimilates will depend on its competitive ability. If hormones are involved in the transfer of assimilates from the phloem to the sink and in the growth of the sink itself, then either or both of these processes might be limited by low levels of endogenous hormones (Wareing, 1978).

Applications of auxin; (which is thought to play a role in the rate of unloading of assimilates from the phloem (Wareing, 1978)), in combination with cytokinin (which is known to increase cell division (Moore, 1979) and so increase sink size) (Wareing, 1978), directly to the pods via a drop leg sprayer (Chapter 6), showed some increase in pod retention especially at the raceme forming pods at the time of application and on those directly below it. Compared to other treatments that did not have the extra plant growth substance application, the overall number of harvestable pods was also increased. When the same treatment was repeated on a field scale (Chapter 7), it again resulted in a similar result at raceme 4 and on those directly below. In turn, this resulted in a significant ($p < 0.001$), increase in the overall percentage of flowers retained to harvestable pods compared to both other treatments and control plants.

This suggests that although the application of auxin and cytokinin did increase sink strength, the effects of the application were short lived, i.e. the plant growth substance application was only able to increase the competitive effect of those pods that were at the correct physiological stage (just set), at the time of application. In order to increase retention until harvest at sites on raceme 5 and above, it can be suggested that further applications would need to be made at for example, growth stages 61 and 81.

Aufhammer (1990), states that there is a correlation between dry matter accumulation in grain legumes and concentrations of active GA and IAA. It may follow therefore, that in addition to the requirements of cytokinin and auxin, GAs are also required within the developing pod to induce assimilate importation. The use of plant growth retardants, which appear to affect the levels of gibberellin synthesis until after the end of flowering (Chapter 5), may therefore be partly responsible for reduced growth of the established but "inactive" pods. It is possible therefore, that applications of extra GAs at the growth stages mentioned above

(41, 61, 81), may also be necessary. Thus, in order to determine which hormones are required to successfully improve sink strength in *Vicia faba* within a field situation, experiments involving the spraying of different plant growth substances and/or combinations of these plant growth substances to the pods at regular intervals should be conducted.

It was noted in Chapter 7, that although the application of extra plant growth substances at growth stage 41 increased the number of harvestable pods compared to a single application of EL500, the overall yield of seed was unaltered (Tables 7.8, 7.10). Similar results were obtained in Chapter 6, where the number of harvestable pods was increased due to an extra plant growth substance application, but yield of seeds was comparable to both single applications of plant growth substances and control plants (Table 6.3). This suggests that although the problems of sink limitation can be overcome by plant growth regulator application, source limitation may then be a factor.

A list of factors that may contribute to larger dry matter yields, include the amount of light intercepted, photosynthetic rate and the capacity of the plant to partition assimilates into specific plant organs (Slinkard and Sindhu, 1988). Increasing plant population results in the mutual shading of plants (Hodgeson and Blackman, 1957; Smith, 1982). Thus, although the actual amount of photosynthetically active radiation (PAR), intercepted per unit area may be the same, that intercepted by individual plants may be reduced (Dantuma and Thompson, 1983). Indeed (see Chapter 7), as density is increased, the number of pods per plant is reduced. However, as density increases, yield per unit area increases (Chapter 7, discussion). Thus, the correct plant population for maximum total yield per unit area must be established. Field trials (Chapter 7), suggest that for Threefold White, this population may be 30 plants m^{-2} in controls, but 40 plants m^{-2} after treatment with plant growth substances. Leaf area per plant is also an important component of light interception

and photosynthetic capacity. In Chapter 5, however, it was shown that the proportion of total dry matter contained in the leaves was generally unaffected by the application of growth retardants and in some cases was actually increased. Therefore, it can be suggested that any source limitation is not due to a reduction in leaf area caused by plant growth substance application.

Photosynthetic rate of the crop can be measured as the amount of dry matter produced within the crop in relation to the photosynthetically available radiation (PAR), Hay and Walker (1989). In Maris Bead, over a 30 day period, growth rates of 30g m^{-2} per day, corresponded to a conversion rate of 3.4% (Fasheun and Dennett, 1982), a very high rate for C_3 species (Dantuma and Thompson, 1983). It is highly unlikely therefore, that breeding techniques could significantly increase this rate (Dantuma and Thompson, 1983). Indeed, if yield can be considered to be a result of this photosynthetic efficiency, when high yielding modern cultivars are compared with ancestral lower yielding ones, there are no appreciable differences in photosynthetic rate (Evans, 1984).

The efficiency of conversion of photosynthate to seed storage material can be expressed as the harvest index of the plant. (Hay and Walker, 1989). It can be seen from Tables 3.5 and 7.11, that the application of plant growth substances can increase the harvest index of the plant. Comparisons of Tables 7.10 and 7.11, show that as this harvest index increased, the yield of beans increased. Harvest index was increased in chapter 7, from 19.% in control plants to 27% in all treatments. In addition, the yield of treatments was also similar. This suggests that: 1. unless more growth regulator applications are made at growth stages 61 and 81, (thus, increasing sink capacity still further), then the harvest index and as a consequence yield may reach a plateau. or 2. Source limitations play a significant role after the attainment of a harvest index of 27%. Thus, there may be a finite limit to the amount of assimilates produced by the plant, with this limit being approached after

the application of plant growth substances. The presence or not of this finite limit cannot be known until sink capacity is determined by experiments applying more plant growth substances at growth stages previously mentioned have been performed. Yields of up to 13t/ha dry matter have been recorded (Fasheun and Dennett, 1982), but of this dry matter only 3.5t/ha was in the form of seed yield which represents a harvest index of 21%. This is therefore comparable to harvest indices of control plants in Chapter 7. It is unknown what the maximum harvest index in *Vicia faba* could be and is unlikely to be known until the experiments suggested are performed. However, an ultimate goal must be to try and achieve an harvest index of approx. 50% as can be achieved in cereals.

In conclusion, it is possible to overcome the sink limitations of *Vicia faba*, caused by premature flower drop, by the application of plant growth substances. With single applications, increased yield potential due to this increased pod set is, however, generally unrealised. A second application of plant growth substances was shown to increase the competitive effect of these pods. It may be possible therefore, to improve these effects by further applications of plant growth substances (e.g. auxins, cytokinins or gibberellins). With such a potentially large increase in sink capacity, it may follow that a second problem of source limitation could exist. Applications of nitrogen with extra applications of plant growth substances and at different densities appeared to have little effect on overall assimilate production. Thus, although the number of harvestable pods per plant can be increased and the assimilate partitioning to these pods improved by the application of plant growth substances, until a reliable method of increasing dry matter production is achieved, faba beans will remain sensitive to the field environment.

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Appendix A.14: Effect of plant growth regulators on root elongation rate

Appendices for Chapter 3

Treatment	Plasma Frequency					
	1	2	3	4	5	6
Control	25.6	33.6	6.3	1.7	1.3	0.0
A	25.6	29.2	10.3	12.3	1.3	0.7
B	24.2	25.8	7.5	5.3	0.0	0.0
C	42.3	22.8	21.3	9.9	1.3	0.3
D	51.3	32.9	30.1	16.5	4.6	1.0
E	55.2	49.1	26.3	17.7	3.1	1.3
F	40.7	35.1	19.8	8.9	1.5	1.0
G	62.5	55.2	40.7	25.3	6.4	3.4
H	50.4	54.0	16.9	2.8	1.6	1.3
I	33.8	36.5	20.8	15.4	2.2	0.8
J	62.3	67.4	31.2	23.2	11.5	10.8
K	17.1	29.0	33.3	11.3	1.0	0.0
L	66.3	55.0	37.3	18.0	2.5	1.1
SED (1/8 df)	3.87	6.27	7.25	8.77	3.02	2.19
t-value	<0.001	<0.001	<0.001	<0.001	0.012	<0.001

Appendix 3.1: Effect of plant growth retardants on intra-raceme pod set

Treatment	Flower Position					
	1	2	3	4	5	6
Control	54.6	33.6	6.5	1.7	1.0	0.0
A	60.0	50.2	19.3	13.5	1.3	0.7
B	47.2	25.9	7.5	2.1	0.0	0.0
C	62.8	52.6	21.9	9.4	1.3	0.0
D	61.2	59.9	30.1	16.5	4.6	1.9
E	53.2	40.7	26.3	12.2	2.1	1.3
F	70.7	55.1	19.8	9.3	1.6	1.0
G	62.5	55.9	40.7	32.3	6.4	3.5
H	70.4	54.0	16.9	5.5	3.6	1.8
I	69.8	54.5	28.3	15.5	2.2	0.0
J	68.3	67.4	50.2	33.8	12.5	10.8
K	74.1	59.0	23.5	15.3	3.0	0.0
L	66.8	55.6	27.5	16.8	2.6	1.1
SED (178 df)	5.87	6.91	7.28	6.17	3.06	2.10
f value	<0.001	<0.001	<0.001	<0.001	0.012	<0.001

Appendix 3.3: Effect of plant growth retardants on inter-raceme pod set

Treatment	Raceme Number						
	1	2	3	4	5	6	7
Control	25.0	27.4	25.1	22.3	15.5	13.7	7.7
A	39.4	34.5	33.1	33.0	30.0	31.9	19.6
B	21.0	22.3	27.8	23.1	18.6	8.6	2.4
C	35.0	29.8	33.5	30.4	23.5	25.4	15.3
D	38.4	41.0	43.9	44.8	33.5	30.4	22.0
E	31.6	31.7	40.5	35.9	30.7	26.6	14.6
F	35.2	29.5	30.4	34.8	33.0	27.4	21.7
G	52.6	47.6	45.9	42.2	36.4	28.4	25.3
H	37.5	32.9	29.2	34.3	29.7	28.7	23.0
I	33.3	39.2	33.5	39.0	26.8	29.3	21.7
J	47.6	42.8	44.6	55.4	52.3	44.4	37.3
K	31.2	36.9	38.7	30.9	29.0	35.5	34.3
L	35.0	31.3	31.0	39.7	30.5	27.2	20.1
SED (178 df)	7.33	5.63	5.34	5.11	5.39	6.58	6.63
f value	0.004	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

Appendix 3.3: Effect of plant growth retardants on intra-raceme percentage mature pods

Treatment	Flower Position					
	1	2	3	4	5	6
Control	43.8	8.8	0.0	0.0	0.0	0.0
A	47.9	17.3	4.6	1.3	0.0	0.0
B	34.0	5.3	2.8	0.7	0.0	0.0
C	46.5	15.4	0.7	0.0	0.0	0.0
D	45.1	28.8	4.9	2.2	0.0	0.0
E	40.0	18.2	2.6	0.0	0.0	0.0
F	43.6	11.8	2.3	0.0	0.0	0.0
G	34.2	27.2	14.1	0.7	0.0	0.0
H	44.1	16.2	0.6	0.0	0.8	0.0
I	43.8	18.6	7.3	0.8	0.0	0.0
J	44.0	24.3	15.4	4.1	2.4	0.0
K	52.1	14.7	0.8	0.0	0.0	0.0
L	48.1	25.1	5.7	0.0	1.1	0.0
SED (178 df)	4.77	4.99	2.81	1.11	0.86	0.00
f value	0.010	<0.001	<0.001	0.008	0.142	0.142

Appendix 3.4: Effect of plant growth regulators on inter-raceme percentage mature pods

Treatment	Raceme Number						
	1	2	3	4	5	6	7
Control	19.4	18.6	12.8	15.0	6.7	4.4	0.0
A	29.3	29.2	22.9	14.4	14.1	9.0	4.5
B	17.1	16.8	15.4	11.0	4.6	2.7	0.0
C	24.2	22.9	20.4	14.8	8.1	5.4	1.9
D	26.1	29.1	21.2	21.6	14.9	10.5	5.5
E	26.9	21.0	18.4	18.3	10.0	10.3	4.8
F	26.2	22.5	19.3	13.6	6.3	2.7	1.9
G	35.0	33.0	22.3	14.8	11.7	5.9	4.3
H	27.1	24.0	16.9	12.9	8.7	8.0	3.8
I	26.2	28.1	21.5	14.0	6.2	2.5	3.9
J	30.9	25.0	24.9	23.5	17.0	11.7	9.4
K	18.4	19.3	18.2	14.9	10.6	9.8	6.8
L	25.9	24.1	19.2	15.3	13.6	7.7	4.9
SED (178 df)	6.52	4.53	3.61	3.54	3.83	3.15	3.05
f value	0.302	0.015	0.086	0.034	0.024	0.018	0.139

Appendix 3.5: Effect of plant growth retardants on plant height

Treatment	Vegetative Height (cm)	Reproductive Height (cm)
Control	40.6	108.4
A	31.4	77.8
B	31.0	96.8
C	35.8	98.5
D	41.3	56.5
E	39.2	87.7
F	39.0	92.6
G	37.1	61.5
H	39.5	87.9
I	43.4	90.4
J	37.2	62.2
K	46.4	88.6
L	41.3	93.6
SED (178 df)	2.37	4.55
f value	<0.001	<0.001

Appendix 4.1: Effect of applied plant growth regulators on root growth of *Pinus pinaster* (mm) means per average pool set

Appendices for Chapter 4

Treatment	Treatments							
	1	2	3	4	5	6	7	8
Control	15	16	17	18	19	20	21	22
A	17	18	19	20	21	22	23	24
B	19	20	21	22	23	24	25	26
C	21	22	23	24	25	26	27	28
D	23	24	25	26	27	28	29	30
E	25	26	27	28	29	30	31	32
F	27	28	29	30	31	32	33	34
SE (24 df)	0.6	0.7	0.8	0.9	1.0	1.1	1.2	1.3
F value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

Appendix 4.2: Effect of applied plant growth regulators on root growth of *Pinus pinaster* (mm) means per average pool set

Replicate Number	Treatments							SE (24 df)	F value
	Control	A	B	C	D	E	F		
1	21	1	22	11	31	42	14	18.1	<0.001
2	14	6	22	29	11	17	1	1.1	<0.001
3	9	5	17	10	23	98	96	7.6	<0.001
4	20	13	15	10	34	102	95	4.1	<0.001
5	14	16	1	13	22	10	103	4.1	<0.001
6	9	6	18	17	23	94	96	6.7	<0.001
7	11	6	18	13	16	93	91	3.3	<0.001
8	10	9	12	9	22	85	105	4.7	<0.001
9	9	13	12	10	15	91	90	7.9	<0.001
10	5	16	17	5	14	91	89	1.7	<0.001
11	1	21	4	1	29	81	96	10.4	<0.001
12	5	17	18	1	42	98	92	9.7	<0.001
13	4	13	6	3	27	92	89	11.5	<0.001
14	4	21	7	29	25	92	89	24.2	<0.001

Appendix 4.1: Effect of applied plant growth substances to each flower on intra-raceme percentage pod set

Treatment	Flower Position							
	1	2	3	4	5	6	7	8
Control	35	19	15	4	0	4	4	0
A	37	24	6	7	1	0	3	0
B	56	17	13	3	0	1	0	0
C	46	14	7	5	4	4	1	3
D	76	44	16	16	6	3	2	0
E	98	98	97	91	96	95	92	97
F	93	92	90	87	86	85	81	90
SED (24 df)	8.6	8.2	7.2	5.7	4.2	4.3	4.4	3.1
f value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

Appendix 4.2: Effect of applied plant growth substances to each flower on the inter-raceme percentage pod set

Raceme Number	Treatment							SED (24 df)	f value
	Control	A	B	C	D	E	F		
1	21	4	22	17	52	100	90	14.3	<0.001
2	14	6	22	20	41	92	94	9.3	<0.001
3	9	5	17	10	33	97	96	7.8	<0.001
4	20	12	15	10	34	100	95	6.7	<0.001
5	14	16	8	13	22	98	100	4.2	<0.001
6	9	6	18	17	21	98	95	6.7	<0.001
7	11	6	14	13	16	95	98	5.3	<0.001
8	10	9	12	9	22	95	93	6.7	<0.001
9	9	13	15	10	19	92	90	7.9	<0.001
10	3	16	17	5	14	91	68	9.7	<0.001
11	1	21	4	5	26	91	76	11.6	<0.001
12	5	17	18	8	47	96	90	9.7	<0.001
13	4	15	6	5	37	92	80	11.6	<0.001
14	4	21	7	29	25	80	60	24.0	<0.001

Appendix 4.3: Effect of applied plant growth substances to each flower on intra-raceme percentage mature pods

Treatment	Flower Position							
	1	2	3	4	5	6	7	8
Control	18	8	4	2	0	1	0	0
A	18	9	1	0	1	0	0	0
B	37	4	1	0	0	0	0	0
C	24	1	3	1	3	0	0	0
D	29	9	0	4	0	0	0	0
E	25	4	4	4	9	4	9	3
F	15	4	3	7	14	4	4	5
SED (24 df)	8.5	3.3	2.0	2.3	3.6	2.1	3.4	2.8
f value	0.191	0.118	0.337	0.064	0.003	0.199	0.104	0.250

Appendix 4.4: Effect of applied plant growth substances to each flower on the inter-raceme percentage mature pods

Raceme Number	Treatment							SED (24 df)	f value
	Control	A	B	C	D	E	F		
1	7	4	5	10	25	20	15	14.5	0.720
2	8	3	8	16	14	3	13	4.8	0.058
3	5	2	6	6	8	2	6	4.2	0.723
4	10	5	10	5	11	12	13	4.3	0.384
5	4	4	7	8	5	9	5	3.4	0.605
6	4	3	10	5	3	10	12	5.4	0.501
7	5	0	8	7	5	10	10	3.9	0.172
8	0	2	5	2	0	3	5	3.4	0.646
9	3	3	16	0	0	9	5	3.4	0.001
10	2	8	8	0	7	11	3	6.1	0.548
11	0	3	3	3	3	8	3	4.5	0.680
12	0	8	8	3	17	7	0	7.1	0.258
13	2	5	3	0	7	26	0	11.3	0.291
14	5	8	3	3	0	10	11	6.5	0.618

Appendix 4.5: Effect of applied plant growth substances to each flower on the reproductive height.

Treatment	Reproductive Height (cm)
Control	111.7
A	128.8
B	124.2
C	134.0
D	102.8
E	120.2
F	106.6
SED (24 df)	14.92
f value	0.332

Appendix 5.1: Effect of Ahr, IF 10405 and HL500 on water-uptake, (Continued) part 2: at growth stage 91

Appendices for Chapter 5

Treatment	Harvest Position						
	1	2	3	4	5	6	7
Control	41.5	37.5	19.5	7.0	3.0	5.0	2.5
Ahr	65.5	29.5	10.5	7.5	4.5	9.0	5.2
IF 10405	67.5	32.5	17.5	16.4	10.5	9.0	3.5
HL500	25.5	30.5	35.0	19.5	9.7	5.7	7.5
SED (11.40)	17.61	12.56	8.29	8.34	4.95	4.90	4.25
F value	0.418	0.587	0.631	0.438	0.5	0.418	1.878

Appendix 5.2: Effect of Ahr, IF 10405 and HL500 on water-uptake, (Continued) part 3: at growth stage 91

Treatment	Harvest Position							
	1	2	3	4	5	6	7	8
Control	12.5	7.5	9.4	6.5	11.1	5.7	14.5	12.4
Ahr	17.3	20.7	13.9	15.4	11.8	10.7	23.0	15.2
IF 10405	21.3	32.3	29.5	30.7	27.3	30.7	34.3	25.8
HL500	31.1	28.9	24.5	30.5	25.2	34.5	29.8	24.9
SED (12.40)	7.49	4.98	7.65	7.66	5.51	8.45	4.27	7.58
F value	0.433	0.084	0.577	0.097	0.405	0.023	0.239	0.406

Appendix 5.1: Effect of Alar, JF 10405 and EL500 on intra-raceme percentage pod set measured at growth stage 91.

Treatment	Flower Position						
	1	2	3	4	5	6	7
Control	47.5	27.5	10.0	5.0	5.0	5.0	2.5
Alar	62.5	20.0	10.0	7.5	8.6	0.0	6.2
JF 10405	87.5	52.5	17.5	26.4	12.5	0.0	3.3
EL500	87.5	50.0	35.0	13.2	5.7	5.7	2.9
SED (12 df)	17.98	12.56	8.20	8.34	8.86	5.59	4.38
f value	0.118	0.057	0.031	0.098	0.828	0.616	0.828

Appendix 5.2: Effect of Alar, JF 10405 and EL500 on inter-raceme percentage pod set at growth stage 91.

Treatment	Raceme Number							
	1	2	3	4	5	6	7	8
Control	12.6	9.3	9.4	9.5	11.1	8.7	18.8	15.4
Alar	17.3	20.7	13.9	16.4	21.8	10.7	20.0	15.4
JF 10405	21.3	33.3	29.5	30.7	37.1	30.7	34.3	23.8
EL500	31.3	28.9	24.3	30.7	35.0	34.6	29.6	24.6
SED (12 df)	7.49	8.88	7.65	7.60	8.61	8.85	8.27	7.26
f value	0.133	0.084	0.077	0.037	0.035	0.023	0.230	0.426

Appendix 5.3: Effect of Alar, JF 10405 and EL500 on intra-raceme percentage pod set, measured at the end of flowering.

Treatment	Flower Position						
	1	2	3	4	5	6	7
Control	45.2	27.4	18.9	18.2	4.2	9.0	2.4
Alar	60.6	42.5	15.3	3.6	4.7	4.9	0.0
JF 10405	56.1	34.6	28.6	26.2	6.7	13.2	16.0
EL500	48.5	27.9	17.2	8.3	11.9	3.1	0.0
SED (12 df)	10.50	7.90	9.51	9.42	3.85	5.30	4.47
f value	0.472	0.240	0.531	0.129	0.228	0.278	0.010

Appendix 5.4: Effect of Alar, JF 10405 and EL500 on inter-raceme percentage pod set, measured at the end of flowering.

Appendix 5.4: Effect of Alar, JF 10405 and EL500 on inter-raceme percentage pod set, measured at the end of flowering.

Treatment	Raceme Number									
	1	2	3	4	5	6	7	8	9	10
Control	3.3	10.2	18.9	9.9	21.1	21.9	20.3	14.7	16.8	24.5
Alar	20.0	24.6	24.8	35.6	23.6	21.5	27.4	24.4	18.9	17.7
JF 10405	34.5	20.9	25.3	23.9	29.3	36.0	41.7	28.1	15.2	21.1
EL500	17.3	22.3	40.8	27.3	29.8	28.1	18.1	20.4	7.3	7.3
SED (12 df)	11.46	8.34	10.11	9.25	10.67	8.03	6.59	8.36	6.68	10.33
f value	0.110	0.358	0.217	0.094	0.807	0.285	0.016	0.454	0.369	0.413

Appendix 5.5: Effect of Alar, JF 10405 and EL500 on intra-raceme percentage pod set, measured at harvest.

Treatment	Flower Position						
	1	2	3	4	5	6	7
Control	62.7	27.5	11.8	7.5	7.3	4.4	8.2
Alar	69.9	52.5	19.9	16.4	1.8	10.4	0.0
JF 10405	61.9	42.6	21.8	27.9	13.5	7.3	7.4
EL500	53.9	36.4	21.1	16.3	9.8	10.6	3.5
SED (12 df)	9.88	12.1	5.04	7.11	4.48	5.45	3.32
f value	0.481	0.263	0.216	0.089	0.118	0.632	0.111

Appendix 5.6: Effect of Alar, JF 10405 and EL500 on inter-raceme percentage pod set, measured at harvest.

Treatment	Raceme Number									
	1	2	3	4	5	6	7	8	9	10
Control	22.2	21.3	21.5	17.9	14.4	25.5	26.8	5.0	33.2	44.5
Alar	35.7	24.0	29.2	32.2	34.9	34.7	28.7	20.9	29.7	30.3
JF 10405	33.7	28.1	32.4	35.9	43.6	45.2	37.9	38.5	30.9	28.1
EL500	21.7	16.6	34.1	21.2	46.1	35.5	33.0	17.7	27.4	7.9
SED (12 df)	12.86	12.75	11.47	8.84	8.67	9.72	6.33	8.16	16.12	14.87
f value	0.589	0.834	0.705	0.181	0.013	0.296	0.343	0.012	0.987	0.159

Appendix 5.7: Effect of Alar, JF 10405 and EL500 on intra-raceme percentage of harvestable pods.

Treatment	Flower Position						
	1	2	3	4	5	6	7
Control	34.6	8.3	2.9	0.0	4.2	3.2	0.0
Alar	28.1	12.2	2.7	0.0	0.0	0.0	0.0
JF 10405	26.3	11.8	3.5	4.4	1.1	0.0	0.0
EL500	23.2	11.3	2.5	1.8	1.5	0.0	0.0
SED (12 df)	6.01	4.85	2.75	1.71	2.61	1.45	0.00
f value	0.329	0.842	0.986	0.072	0.454	0.119	0.00

Appendix 5.8: Effect of Alar, JF 10405 and EL500 on inter-raceme percentage harvestable pods

Treatment	Raceme Number									
	1	2	3	4	5	6	7	8	9	10
Control	10.3	4.0	13.1	11.2	7.5	11.1	11.4	2.5	6.7	13.7
Alar	16.2	21.5	14.8	13.3	13.4	9.4	5.4	2.5	2.5	0.0
JF 10405	4.0	3.3	9.0	17.2	25.0	20.1	9.5	10.0	6.7	0.0
EL500	16.7	8.0	10.2	12.0	8.3	15.3	8.2	5.3	9.5	2.5
SED (12 df)	8.88	5.36	7.39	6.78	5.24	5.46	5.39	5.06	7.88	3.34
f value	0.472	0.018	0.856	0.818	0.021	0.256	0.727	0.435	0.846	0.004

Appendix 5.9: Effect of Alar, JF 10405 and EL500 on dry matter distribution, measured at growth stage 41.

Treatment	Total Weight Dry Matter (g)	% of D.M. in Stem	% of D.M. in Leaves	% of D.M. in Root
Control	7.94	43.89	33.31	22.8
Alar	6.14	43.57	32.53	23.9
JF 10405	6.86	44.79	30.77	24.4
EL500	6.52	40.42	35.53	24.0
SED (12 df)	0.888	2.216	2.203	2.910
f value	0.259	0.271	0.241	0.949

Appendix 5.10: Effect of Alar, JF 10405 and EL500 on dry matter distribution, measured at growth stage 91.

Treatment	Total Weight Dry Matter (g)	% of D.M. in Stem	% of D.M. in Leaves	% of D.M. in Roots	% D.M. in Pods/Beans
Control	10.7	49.21	28.00	20.45	2.37
Alar	9.3	40.31	28.80	25.90	5.02
JF 10405	9.8	40.28	31.70	24.96	3.07
EL500	8.0	38.97	37.90	25.19	5.67
SED (12 df)	1.81	1.63	3.18	2.14	1.90
f value	0.513	<0.001	0.035	0.093	0.302

Appendix 5.11: Effect of Alar, JF 10405 and EL500 on dry matter distribution, measured at the end of flowering.

Treatment	Total Weight of Dry Matter (g)	% of D.M. in Stem	% of D.M. in Leaves	% of D.M. in Root	% of D.M. in Pod/Bean
Control	18.9	42.80	26.41	18.90	11.90
Alar	22.5	32.00	23.14	20.50	24.30
JF 10405	22.3	35.00	30.18	20.90	13.90
EL500	17.7	36.20	25.97	20.50	17.30
SED (12 df)	4.29	3.03	2.52	3.24	3.67
f value	0.606	0.024	0.097	0.932	0.025

Appendix 5.12: Effect of Alar, JF 10405 and EL500 on plant height, measured at growth stage 91.

Treatment	Vegetative Height (cm)	Reproductive Height (cm)
Control	33.2	71.0
Alar	43.6	59.2.
JF 10405	35.6	54.2
EL500	35.6	47.6
SED (12 df)	4.11	3.40
f value	0.115	<0.001

Appendix 5.13: Effect of Alar, JF 10405 and EL500 on plant height, measured ^{at the} end of flowering.

Treatment	Vegetative Height (cm)	Reproductive Height (cm)
Control	39.6	98.4
Alar	41.6	72.2
JF 10405	38.4	83.8
EL500	35.2	80.0
SED (12 df)	5.69	6.49
f value	0.725	0.011

Appendix 5.14: Effect of Alar, JF 10405 and EL500 on plant height, measured at harvest.

Treatment	Vegetative Height (cm)	Reproductive Height (cm)
Control	38.2	108.8
Alar	38.0	72.6
JF 10405	37.2	84.8
EL500	35.0	77.4
SED (12 df)	2.55	13.59
F value	0.592	0.086

Appendix 6.1: Effect of FLNRI, DAP and CLIAA, using conventional and deep-leg spraying techniques on *Myndus* spp. (%)

Appendices for Chapter 6

Treatment	Flower Position						
	1	2	3	4	5	6	7
Control	43.5	36.9	31.2	10.3	7.9	3.3	4.3
A	58.1	34.9	25.3	17.2	11.2	23.4	3.5
B	37.9	34.9	18.3	20.9	12.1	6.8	8.4
C	44.8	47.3	36.1	4.2	37.9	23.1	31.3
D	60.8	46.4	36.0	37.0	42.7	39.6	33.3
E	59.0	47.7	34.0	42.0	37.9	36.9	27.9
F	56.3	48.2	40.5	36.2	26.0	23.3	29.3
SSD (24 df)	10.36	2.81	19.27	11.53	14.54	14.39	7.17
F value	0.207	0.036	0.005	0.030	0.010	0.110	<0.001

Appendix 6.2: Effect of FLNRI, DAP and CLIAA, using conventional and deep-leg spraying techniques on *Myndus* spp. (%)

Treatment	Flower Position								
	1	2	3	4	5	6	7	8	9
Control	19.7	45.4	11.2	7.6	7.8	12.8	10.9	17.3	20.5
A	59.0	23.7	26.9	30.9	18.5	11.2	21.3	24.1	29.3
B	34.3	42.1	17.6	3.8	12.8	18.9	13.4	13.1	33.7
C	100.0	34.2	26.9	42.5	10.8	76.8	39.3	37.9	31.8
D	62.6	37.5	31.8	19.5	11.3	13.9	10.8	34.1	30.6
E	41.3	36.7	12.4	10.4	18.7	34.1	10.7	10.8	45.8
F	71.3	27.0	26.1	11.3	17.9	48.3	34.8	31.1	37.9
SSD (24 df)	16.34	18.97	16.13	12.45	18.79	12.39	12.35	12.35	10.22
F value	<0.001	0.002	0.007	0.107	0.005	<0.001	<0.001	0.001	<0.001

Appendix 6.1: Effect of EL500, BAP and CLIAA, using conventional and drop-leg spray techniques on intra-raceme pod set.

Treatment	Flower Position						
	1	2	3	4	5	6	7
Control	45.5	26.9	11.9	10.1	2.9	1.5	4.2
A	58.1	34.9	25.5	17.2	11.2	22.4	3.5
B	37.5	24.3	18.3	20.6	12.1	6.8	8.4
C	44.9	47.3	36.1	34.7	37.9	27.1	31.3
D	60.8	46.4	36.0	37.0	41.7	39.6	33.3
E	59.8	47.7	54.0	47.0	37.9	36.0	27.9
F	56.3	46.2	40.5	38.2	26.0	25.3	29.3
SED (24 df)	10.36	8.81	13.27	11.53	11.54	14.19	7.17
f value	0.207	0.038	0.066	0.039	0.010	0.110	<0.001

Appendix 6.2: Effect of EL500, BAP and CLIAA using conventional and drop-leg spraying techniques on inter-raceme pod set.

Treatment	Raceme Number								
	1	2	3	4	5	6	7	8	9
Control	19.7	15.4	11.2	7.6	7.9	15.3	16.8	15.3	20.0
A	10.0	23.1	26.9	20.7	28.3	11.0	22.3	28.1	29.2
B	53.3	47.1	17.0	7.9	11.3	18.3	18.4	13.1	12.7
C	100.0	55.2	50.6	42.5	65.0	76.1	59.5	37.2	21.8
D	53.0	57.5	31.4	32.5	32.5	58.6	81.5	69.2	70.6
E	93.3	76.7	52.0	44.4	35.7	71.1	62.7	50.5	28.0
F	71.3	57.0	36.1	41.1	57.5	49.2	50.0	51.1	35.3
SED (24df)	16.56	18.57	15.33	16.85	18.38	12.98	10.36	12.35	10.32
f value	<0.001	0.042	0.097	0.138	0.037	<0.001	<0.001	0.001	<0.001

Appendix 6.3: Effect of EL500, BAP and CLIAA, using conventional and drop-leg spray techniques on intra-raceme percentage harvestable pods.

Treatment	Flower Position						
	1	2	3	4	5	6	7
Control	33.2	14.9	3.8	4.3	1.5	0.0	0.0
A	17.2	8.9	4.1	2.2	4.0	0.0	0.0
B	18.9	9.7	4.5	9.9	4.8	1.7	4.2
C	18.6	13.0	7.0	5.3	5.4	0.0	2.9
D	23.6	7.3	8.7	9.1	9.0	3.9	6.3
E	26.0	23.1	17.2	10.4	2.0	3.9	0.0
F	23.7	12.7	4.6	4.7	1.4	4.3	5.5
SED (24 df)	6.79	6.46	4.75	5.79	3.12	2.63	3.90
f value	0.280	0.286	0.101	0.718	0.216	0.216	0.445

Appendix 6.4: Effect of EL500, BAP and CLIAA with conventional and drop-leg spraying techniques on percentage inter-raceme harvestable pods.

Treatment	Raceme Number								
	1	2	3	4	5	6	7	8	9
Control	19.7	9.7	11.2	5.6	4.2	6.8	7.0	10.2	20.0
A	0.0	6.2	10.7	10.4	20.8	2.5	5.6	18.7	11.7
B	3.3	15.7	13.7	7.9	7.3	8.9	11.1	13.1	4.9
C	23.3	10.0	7.5	6.9	7.5	16.9	11.4	11.4	6.9
D	0.0	8.0	2.5	5.0	7.2	21.7	13.8	14.7	16.1
E	28.0	18.0	30.4	20.6	10.4	13.4	10.2	13.9	11.5
F	0.0	13.7	11.1	11.3	15.8	18.7	6.7	6.9	9.3
SED (24df)	10.20	7.93	5.69	7.21	8.08	8.54	6.39	7.18	6.52
f value	0.025	0.738	0.003	0.390	0.422	0.287	0.838	0.775	0.304

Appendix 6.5: Effect of EL500, EL500 and BAP and BAP and CLIAA using conventional and drop leg spray techniques

Treatment	Vegetative Height (cm)	Reproductive Height (cm)
Control	30.0	110.6
A	31.8	61.6
B	30.2	77.4
C	30.2	44.6
D	28.4	55.0
E	31.6	50.8
F	32.8	57.2
SED (24 df)	2.49	8.17
f value	0.662	<0.001

Appendix 8.1: Effect of *Appendices for Chapter 8* (Percentages are in parentheses)

Treatment	Number of Correct Responses				
	1	2	3	4	5
A	14.7	15.7	13.4	4.8	0.0
B	51.0	44.1	20.0	15.4	1.3
C	45.9	47.4	40.1	20.7	18.3
D	91.0	83.3	73.2	31.1	29.2
SED (11.40)	2.31	7.12	4.95	11.39	11.53
F-value	<0.001	<0.001	<0.001	<0.001	<0.001

Appendix 8.2: Effect of *Appendices for Chapter 8* (Percentages are in parentheses)

Treatment	Number of Correct Responses						
	1	2	3	4	5	6	7
A	0.7	0.0	11.1	14.1	2.0	15.0	16.5
B	20.1	21.0	21.4	23.1	22.1	21.0	21.7
C	14.5	25.7	27.3	44.3	32.4	25.3	29.5
D	55.5	54.0	56.7	52.5	48.0	27.2	100.0
SED (11.40)	12.73	7.48	12.47	14.25	16.91	19.81	21.53
F-value	0.001	<0.001	<0.001	0.005	<0.001	0.001	<0.001

Appendix 8.1: Effect of hand-tripping and the application BAP on intra-raceme percentage pod set

Treatment	Flower Position				
	1	2	3	4	5
A	24.7	10.7	1.4	4.6	0.0
B	63.6	44.3	20.3	15.4	3.3
C	65.9	47.4	40.1	30.7	38.5
D	93.0	82.5	73.2	81.1	85.2
SED (11 d.f)	8.91	8.18	8.95	12.38	11.33
f value	<0.001	<0.001	<0.001	<0.001	<0.001

Appendix 8.2: Effect of hand-tripping and the application of BAP on inter-raceme percentage pod set.

Treatment	Raceme Number						
	1	2	3	4	5	6	7
A	0.0	0.0	11.3	14.7	2.9	15.0	19.5
B	20.3	21.9	21.4	23.5	23.2	33.7	43.7
C	14.7	28.7	37.3	41.3	52.4	73.3	79.8
D	58.3	88.0	96.7	80.5	85.0	77.2	100.0
SED (11 df)	12.25	7.48	12.37	14.56	15.03	19.48	13.85
f value	0.003	<0.001	<0.001	0.003	<0.001	0.021	<0.001

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Effects of applied plant growth substances on pod set in broad beans (*Vicia faba* var. *major*)

P. D. RYLOTT AND M. L. SMITH

Edinburgh School of Agriculture, West Mains Road, Edinburgh EH9 3JG, UK

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SUMMARY

The effects on flower drop and pod set of applications of either the auxin 4-chloroindole, the cytokinin 6-benzylaminopurine (BAP) or gibberellic acid (GA_3) to every flower, 1 day before or 1 day after tripping, were studied in broad bean plants grown in a glasshouse in 1988. Control plants shed most of their flowers; pods that did set originated primarily from flowers at proximal positions on the raceme. Most pods set on the lower part of the reproductive portion of the control plants. Chloroindole and GA_3 application before tripping had no significant effect on pod-set pattern. However, GA_3 applied after tripping significantly enhanced pod set at proximal flowers by 21% but inter-raceme pod set was not significantly altered. Chloroindole applied after tripping significantly increased pod set, particularly at proximal flower positions on the raceme; this treatment increased inter-raceme pod set but the increase was significant only for the basal four racemes and racemes 11, 12 and 13. Application of BAP before or after tripping resulted in almost complete pod set on all racemes; but greater flower drop was observed on the uppermost five racemes of plants treated after tripping than on corresponding racemes of plants treated before tripping. The effects of BAP, chloroindole or GA_3 treatment were not due to changes in the synchrony of flower opening. An adequate supply of a cytokinin appeared to be necessary at or before pollination to initiate changes in reproductive organs to prepare them to become net attractors of assimilates from other plant parts. Auxin and gibberellin were only effective in promoting fruit set 24 h after tripping. An adequate supply of auxin, and perhaps of gibberellin, in balance with cytokinin appears to be required after pollination to maintain the flow of adequate assimilates to enable further pod development and for maturation to proceed.

INTRODUCTION

Yield fluctuation in *Vicia faba* is due primarily to reproductive failure which can occur as a result of bud abortion, flower shedding or pod or ovule abortion (Gates *et al.* 1983*b*). Flower drop, which accounts for the major proportion of total reproductive loss, contributes most to the reduction in potential yield. However, abscission within each raceme occurs in most cultivars of *V. faba* in a characteristic pattern. The incidence of shedding at proximal positions is low and stable whereas at more-apical positions it is high and variable (Gates *et al.* 1983*a*). Most European cultivars of *V. faba* possess sequentially opening flowers in each axillary inflorescence. It has been suggested that the pollination of the earliest flower to reach anthesis promotes physiological changes which stimulate pod set (Gates *et al.* 1981). Removal of the proximal two or three flowers on every raceme drastically reduces flower shedding at more-distal positions on the raceme (Smith 1982).

The nature of the abscission promoter translocated from young proximal pods to distal buds is unknown in *V. faba*, although in other legumes indole acetic acid or abscisic acid (ABA) are thought to be involved (Porter 1977; Huff & Dybing 1980).

Experiments using plant growth retardants (Smith 1982; Keller & Belluci 1980; Kellerhals & Keller 1984; Hack *et al.* 1985) have clearly shown that they can significantly increase pod set in *V. faba*. However, the role of endogenous plant hormones in the initiation of pod set and subsequent development of pods, together with the effects that growth regulators (PGRs) may have upon such hormone concentrations, remains unclear. For example, the yield-enhancing effects of application of PGRs to *V. faba* have been variously attributed to a decrease in endogenous concentrations of ABA (El-Zawily *et al.* 1985), or ethylene (Attiya *et al.* 1983). Application of cytokinins in combination with auxin and gibberellic acid to flowers before pollination also results in greater pod set (Chapman & Sadjadi 1981). The present study

investigated the effects of application to flowers, before and after tripping, of an auxin, a gibberellin or a cytokinin, to elucidate the influence of exogenous plant hormones on pod set and flower abscission in broad beans.

MATERIALS AND METHODS

Broad bean, *Vicia faba* var. *major*, plants of the widely grown variety Threefold White were raised in a bee-proof heated glasshouse at the Edinburgh School of Agriculture, Bush Estate in 1988 from sowings of two seeds per 15 cm pot in Fisons Levington potting compost. Seedlings were singled and later staked to prevent lodging. The plants were subject to a daytime minimum temperature of 21 °C, and a minimum night-time temperature of 16 °C. As the experiment was performed in February, 400 W sodium lamps suspended 1 m above the plants, at 1/m², provided a 16 h photoperiod. The experiment was arranged in a randomized block design. Each treatment was replicated five times. Every flower on each plant was treated by gently applying solutions of various plant growth substances on to the standard petal and the calyx, with a fine artist's paintbrush. Three plant hormones were employed: auxin as 4-chloroindole, 3.3×10^{-4} M (50 mg/l); cytokinin as 6-benzylaminopurine (BAP), 1×10^{-4} M (23 mg/l); and gibberellin as GA₃, 2×10^{-5} M (7 mg/l); BAP and GA₃ were first dissolved in a few drops of 1 M sodium hydroxide and then diluted to strength in distilled water containing 3 mg Agral surfactant/l. Chloroindole was dissolved directly in distilled water containing the surfactant. Plants were treated with the growth substance (Table 1) 24 h before or 24 h after pollination. Pollination at the correct time was ensured by hand tripping the flowers. Treatments were related to the time of pollination by hand tripping, as the time to complete fertilization is difficult to determine and can range from 24 to 72 h after pollination (Stoddard 1986). Flowers were judged to be at the correct stage for pollination or hormone treatment when the standard petal and wing petals were unfolding from one another. Anther dehiscence occurs once the standard petal begins to fold back on itself, i.e. stage 9 of the flower development key proposed by Smith (1982).

Flower position within a raceme was assigned a number, 1 being the most-proximal flower, 2 the next and so on. Raceme position was also enumerated: the lowest, and hence first-formed raceme, was numbered 1, and the next 2, until all reproductive nodes had been accounted for. In this way, every flower on the main stem had its own number. This allowed the dates of tripping and chemical application to each flower to be recorded accurately, as well as a detailed analysis of flower and pod drop. The process of pod set was also recorded, and was judged complete once

Table 1. *Treatments and times of application of plant growth substances to Vicia faba*

Treatment	Plant growth substance	Time of application
Control	Distilled water + Agral wetter	24 h before tripping
A	Gibberellic acid	24 h before tripping
B	Gibberellic acid	24 h after tripping
C	4-Chloroindole	24 h before tripping
D	4-Chloroindole	24 h after tripping
E	6-Benzylaminopurine	24 h before tripping
F	6-Benzylaminopurine	24 h after tripping

a young green pod was clearly visible, immediately after petal abscission. Pods were not assessed for parthenocarpy.

To investigate the possible effects of hormone treatment on intra-raceme flower development, a detailed analysis of synchrony of flowering was performed. This was calculated by measurement of the time difference, in days, between the proximal flower having fully opened (stage 9) and the distal flower on the same raceme reaching the same stage of development. This figure was divided by the number of flowers on that raceme. The correlation coefficient was calculated between this intra-raceme flowering synchrony figure and the percentage pod set achieved on the same raceme. This procedure was repeated for every raceme formed for each plant in all treatments.

RESULTS

Pattern of pod set and flower drop

Intra-raceme pod set

In all control plants, the pattern of pod set, and hence that of flower drop, was the same; most pods set at proximal positions on the raceme, especially positions 1 and 2 (Table 2); few pods set from flowers distal to position 3 in the raceme.

Application of GA₃ before tripping (treatment A) resulted in plants that showed a similar pattern of pod setting to that of the controls (Table 2). However, slightly more pods set (5% difference), at position 2 than in the controls but this was counterbalanced by a reduction (10% difference) at position 3. Application of GA₃ after pollination (treatment B) resulted in significantly greater pod set (21% difference) at the proximal flower position on treated plants than on control plants. However, greater pod set on flower position 1 was, compensated for by a reduction of 7% in pod set on flower position 2 compared with treatment A.

The application of chloroindole to flowers 24 h before tripping (treatment C) resulted in an intra-raceme pod set pattern similar to that of control

Table 2. *Effects of application of plant growth substances to each flower on intra-raceme percentage pod set in Vicia faba*

Flower position	Treatment*							S.E.D. (24 D.F.)
	Control	A	B	C	D	E	F	
1 (proximal)	35	37	56	46	76	98	93	8.6
2	19	24	17	14	44	98	92	8.2
3	15	5	13	7	16	97	90	7.2
4	4	7	3	5	16	91	87	5.7
5	0	0	0	4	2	95	86	4.2
6	4	0	1	4	2	95	85	4.3
7	4	2	0	1	2	92	81	4.4
8 (distal)	0	0	0	3	0	97	90	3.0

Data are derived from the mean of five plants per treatment.

* See Table 1.

plants (Table 2); but application after pollination (treatment D) resulted in significantly greater pod set than in control plants at flower positions 1 (41% difference), 2 (25% difference) and 4 (12% difference) (Table 2). The pattern of intra-raceme pod set was maintained, in that most pods set from flowers at proximal positions and fewer pods set at more-distal positions.

The application of BAP to flowers before or after tripping (treatments E and F) resulted in almost complete pod retention and, in both cases, flowers at all raceme positions set significantly more pods ($P < 0.001$) compared with controls and the other treatments (Table 2). Application of BAP before tripping produced 91–98% pod set, whereas application after tripping resulted in less and a greater variation of pod set within racemes, at 81–93%. The pattern of intra-raceme pod set hitherto observed was not apparent; a high percentage pod set was achieved at all flower positions.

Inter-raceme pod set

Control plants set most pods on the first nine reproductive nodes formed (Table 3). Average pod set for each of these nodes was 13%. Pod set declined towards apical reproductive nodes and was an average 3% on each of racemes 9–14.

Flowers brushed with GA_3 before tripping (treatment A) produced an average pod set per raceme for the first nine reproductive nodes of 9% whereas, for the remaining five racemes, 18% was achieved. This was the reverse of the control plants (Table 3). Thus, after treatment A, more pods were set in the upper part of the reproductive portion of these plants, but pod set at any one raceme was not significantly different from that of the control plants. When GA_3 was brushed on to flowers after tripping (Table 3, treatment B), the average pod set for each of the first

Table 3. *Effects of application of plant growth substances to each flower on inter-raceme percentage pod set in Vicia faba*

Raceme number	Treatment*							S.E.D. (24 D.F.)
	Control	A	B	C	D	E	F	
1 (base)	21	4	22	17	52	100	90	14.3
2	14	6	22	20	41	92	94	9.3
3	9	5	17	10	33	97	96	7.8
4	20	12	15	10	34	100	95	6.7
5	14	16	8	13	22	98	100	4.2
6	9	6	18	17	21	98	95	6.7
7	11	6	14	13	16	95	98	5.3
8	10	9	12	9	22	95	93	6.7
9	9	13	15	10	19	92	90	7.9
10	3	16	17	5	14	91	68	9.7
11	1	21	4	5	26	91	76	11.6
12	5	17	18	8	47	96	90	9.7
13	4	15	6	5	37	92	80	11.6
14 (apex)	4	21	7	29	25	80	60	24.0

Data are derived from the mean of five plants per treatment.

* See Table 1.

nine racemes was 16%, whereas for the remaining five racemes it averaged 10%. Again, however, pod set on any one raceme was not significantly different from that achieved by the control plants.

Brushing chloroindole on to flowers before tripping (Table 3, treatment C) resulted in an average 13% pod set on each of the first nine and 10% for the remaining five racemes, but these figures were not significantly different from those for control plants. Chloroindole applied to flowers 24 h after tripping (Table 3, treatment D) increased the average pod set for each of the first nine racemes to 29%; only the most-basal four racemes showed a significant increase compared with that of control plants. The average pod set on the remaining five racemes was 30%. However, pod set was only significantly greater than on control plants on racemes 11, 12 and 13.

The application of BAP to flowers before tripping (Table 3, treatment E) culminated in almost complete pod set on all racemes, with little differentiation between the lower part of the reproductive portion of the plant (96%) and the upper part of the reproductive portion (90%). All racemes had a significantly ($P < 0.001$) greater pod set percentage than on control and other treated plants. Application of BAP to flowers after tripping (Table 3, treatment F) again produced a high pod set, which averaged 95% on each of the first nine racemes, but more flower abscission on the upper five racemes, where pod set averaged 75%. However, this was not significant compared with plants subjected to treatment E, with the exception of raceme 10, which resulted in significantly fewer ($P < 0.05$) pods compared with the corresponding raceme on plants subjected to treatment F.

Overall pod set

Brushing GA₃ onto flowers before or after tripping (Table 4, treatments A and B) had little effect on overall pod set compared with control plants. The application of chloroindole before tripping produced a similar result but, when applied after tripping, it had the significant effect of increasing overall pod set by

Table 4. *Effects of application of plant growth substances to each flower before or after tripping on overall flower drop and pod set in Vicia faba*

Treatment*	Number of flowers per plant	Number of flowers dropped per plant	Number of pods set per plant	Flower drop (%)	Pod set (%)
Control	128.4	114.3	14.0	89.1	10.9
A	118.0	104.0	14.0	88.2	11.8
B	119.6	104.2	15.4	86.9	13.1
C	112.0	99.2	12.8	88.6	11.4
D	85.6	64.2	21.4	74.1	25.9
E	100.6	4.2	96.4	4.1	95.9
F	109.8	12.8	97.0	11.3	88.7
S.E.D. (24 D.F.)	16.47	14.88	5.77	3.95	3.95

Data are the mean of five plants per treatment.

* See Table 1.

Table 5. *Effects of application of plant growth substances to each flower on the intra-raceme synchrony of flowering in Vicia faba; low values indicate more synchrony than high values*

Raceme number	Treatment*							S.E.D. (24 D.F.)
	Control	A	B	C	D	E	F	
1 (base)	0.82	0.83	0.80	0.75	0.85	0.83	0.86	0.154
2	0.58	0.57	0.62	0.51	0.68	0.54	0.61	0.125
3	0.44	0.42	0.51	0.42	0.43	0.39	0.38	0.066
4	0.38	0.40	0.50	0.50	0.38	0.46	0.33	0.062
5	0.38	0.42	0.48	0.43	0.38	0.60	0.38	0.063
6	0.44	0.50	0.43	0.44	0.52	0.52	0.46	0.087
7	0.44	0.43	0.60	0.61	0.61	0.52	0.49	0.086
8	0.53	0.48	0.56	0.48	0.58	0.56	0.42	0.087
9	0.56	0.41	0.60	0.51	0.51	0.61	0.38	0.088
10	0.60	0.65	0.54	0.52	0.80	0.63	0.50	0.158
11	0.48	0.55	0.61	0.61	0.71	0.61	0.66	0.133
12	0.60	0.52	0.60	0.63	0.82	0.74	0.55	0.135
13	0.73	0.56	0.61	0.61	0.84	0.62	0.61	0.152
14 (apex)	0.48	0.78	0.50	0.49	0.64	0.78	0.47	0.140
Correlation coefficient between synchrony and mean pod set on all racemes	-0.05	0.13	0.26	-0.14	0.40	0.32	-0.20	

Data are the mean of five plants per treatment.

* See Table 1.

15% compared with control plants. Application of BAP to plants before or after tripping significantly ($P < 0.001$) increased overall pod set by 85 and 77.8%, respectively (Table 4, treatments E and F).

*Synchrony of flowering**Intra-raceme synchrony of flowering*

Synchrony of flower opening was not generally affected by the application of plant hormones. In only five cases was the synchrony ratio significantly altered and, of these, only one was significantly reduced, i.e. the flowering became more synchronous. In no case was the correlation between the synchrony ratio (Table 5) and the percentage pod set achieved on that raceme (Table 3) $> \pm 0.5$.

Inter-raceme synchrony of flowering

The number of days from the start of flowering until all flowers on raceme 14 had reached the stage at which pollination could take place was not significantly altered by the application of plant growth substances. The length of time in all cases only ranged between 21 and 23 days.

DISCUSSION

Gates *et al.* (1981) suggested that inbred lines that develop few flowers per node and all opening within

2 days are less likely to shed their flowers than those with many flowers per node. In the present experiment, control plants showed no correlation between synchrony of flowering and resulting pod set. In addition, treatments to flowers that did result in increased pod set (C, E and F) resulted in a flowering pattern less synchronous than that of control plants. Kellerhals & Keller (1984) suggested that shortening of the flowering period, associated with application of an anti-gibberellin, may have been partly responsible for the increased pod set associated with that plant growth regulator treatment. However, the application of hormones to flowers in our experiment resulted in no significant change in the length of the flowering period. Therefore, it is suggested that the numbers of pods set in this experiment are a consequence of hormonal action and not due to any effects on the synchrony of flowering. The application of GA_3 before tripping had little effect on pod set. Applied after tripping, it promoted pod set at the proximal flower position. This effect was distributed evenly over all racemes, such that no significant difference was observed in inter-raceme pod set between treated plants and the controls, there being reductions in pod set at other flower positions.

It would be incorrect to attribute the results of the present experiment to the role of endogenous cytokinins. It does appear, however, that exogenous application of cytokinins can promote almost complete pod set and it can be postulated that similar intrinsic chemicals may be extremely important in the process of pod set in *Vicia faba*.

It is well established that cytokinins can promote cell division in the vascular tissue, but that cytokinins are inactive in the absence of auxin (Moore 1979). In this experiment, auxin was presumably present intrinsically, although perhaps in limiting concentrations, so the application of cytokinin to flowers resulted in active cell-division of the embryo, and hence the attraction of assimilates to the now developing pods from other plant parts. This view is supported by results obtained by Chapman & Sadjadi (1981), where benzyl-adenine was applied exogenously to distal flowers only. In their case, treatment of distal flowers caused more pods to set at the expense of untreated proximal ones. Thus, exogenous cytokinins appear to be able to promote the formation of, or perhaps aid the preparation of, potentially active reproductive sinks which are rapidly established after the stimulus of pollination and can then effectively divert resources from competing sinks, primarily the vegetative apex, and from sources such as the leaves and stem.

In peas (*Pisum sativum*), the cytokinins obtained in the pod development phase before embryo growth, resemble those exported from the root system (Burrows & Carr 1970). In *Vicia faba*, lower concentrations of cytokinin may be transported from roots

to other plant parts during flowering, possibly because of reduced root growth during this period. Such reduced root growth may be in part responsible for flower abscission. Lesina (1966) showed that the application of GA_3 reduced the weight of roots in *V. faba*. Consequently, the opposite effect may occur after the application of anti-gibberellin-type plant growth retardants, i.e. an increase in root weight may be observed, increasing in turn cytokinin production so resulting in increased pod set. In addition, Henson & Wheeler (1976) discovered that root nodules in *V. faba* have high intrinsic concentrations of cytokinin. The anti-gibberellin compound cycocel, when applied to field beans (Hassan & El-Moursi 1982), increases nodulation. Enhanced pod set associated with the application of anti-gibberellins in field beans (Richards & Smith 1987) could be partly attributed to an increase in cytokinin concentrations due to greater nodulation.

The change in pattern of flower drop and pod set exhibited by plants supplied with auxin, and to an extent with gibberellin, from before to after tripping (Tables 3, 4), suggests that a 'hormonal switch' occurs in terms of auxin and gibberellin requirement at, or directly after, tripping. Before tripping, reproductive organs are insensitive to auxin and gibberellin application; after tripping, greater pod set results. With regard to auxin, Engvild (1985) showed that chloroindole concentrations in developing field bean seed decreases initially after fertilization. However, in peas, a species that does not suffer from appreciable flower drop (Richards & Smith 1987), no initial decline in chloroindole was observed; instead, concentrations increased by > 10 000 times in the first 20 days after fertilization. Gates *et al.* (1983*b*) stated that in *V. faba* a lag period prior to active pod growth coincided with vascular differentiation at the pedicel-peduncle junction. Auxin is known to play a major role in vascular differentiation (Bandurski & Nonhebel 1984). Thus, the observed increase in pod set displayed by application of chloroindole after tripping can be attributed to the redirection of assimilates primarily to the pedicel-peduncle junction to facilitate vascular differentiation in that region. This role appears to be similar to that of cytokinin, except that cell expansion is probably promoted by auxin application (Moore 1979). This was particularly true of flowers at proximal positions in the raceme, where it may be supposed that cytokinin supply was not limiting.

It appears, however, that both auxin and cytokinin, and to a certain extent gibberellin, in combination have a crucial role in the process of pod set in *V. faba*. It is possible that the stimulus to pod set referred to by Gates *et al.* (1981) may be auxin based. The auxin gradient theory of abscission (Addicott *et al.* 1955) states that abscission is induced when the proximal:distal ratio of auxin across a potential abscission

zone, in this case at the base of the flower, is increased. Diethelm *et al.* (1986) stated that the concentration of hormones within a flower is slightly reduced between bud initiation and full flower development. Thus, if a flower distal within a raceme is considered, it can be suggested that the lowering of internal hormone concentrations, compared with increases in an auxin-based stimulus proximal to it, originating from pollinated flowers and the apex, may induce the cellular processes leading to abscission, whether the flower is fertilized or not. Cytokinins, however, may be more important in the establishment of reproductive sinks, whereas auxins and gibberellins may be involved after fertilization in their maintenance and further development. The results of the present experiment indicate that cytokinins are limiting and that increasing concentrations of these substances in the flower leads to pod set. Hence, limiting supplies of cytokinin and auxin in flowers may lead to the successful formation of an abscission layer at the pedicel-peduncle junction. Further experiments on *V. faba* in the glasshouse and in the field have endeavoured to clarify the effects of timed applications of various plant growth regulators and

plant growth substances. Work is also being conducted into the extent to which BAP may induce parthenocarpic pod set and the effects of applied plant growth regulators on root growth and nodulation.

Kambal *et al.* (1976) stated that not all pollinated flowers develop into mature pods and suggested that factors other than pollination may be important. With evidence from our experiment, it is suggested that this is because hormone concentrations in flowers, at and just after pollination, need to be in a favourable balance. It is apparent that cytokinins need to be present in a favourable ratio before and at pollination, while a supply of auxin and gibberellin must be available immediately afterwards. It might be possible, therefore, to select for genotypes with an altered balance of endogenous hormones. The application of naturally occurring plant hormones at physiological concentrations may be a more acceptable alternative to that of synthetic plant growth regulators.

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